MARCH, 1961
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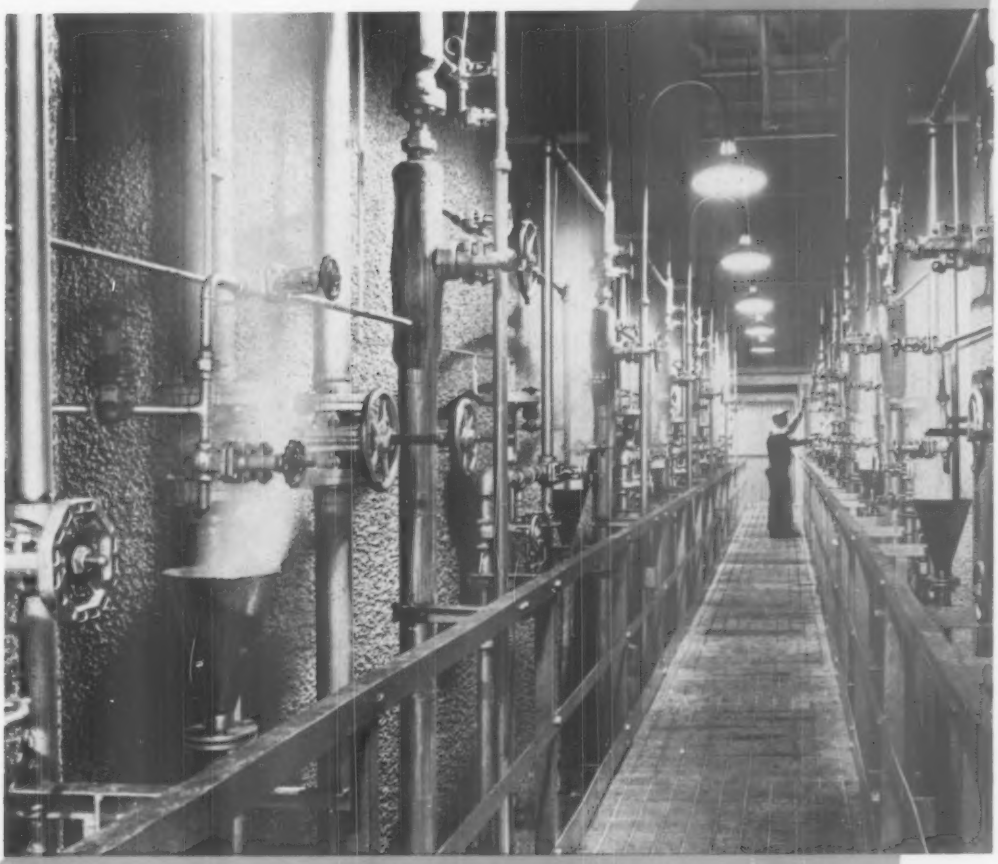
FT food technology



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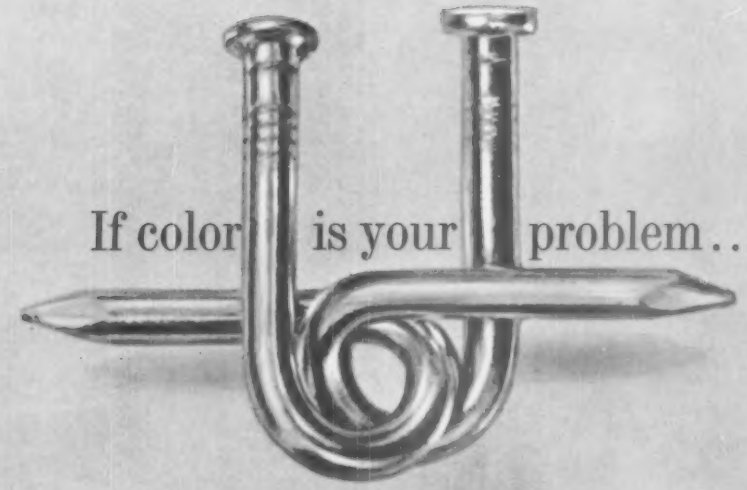

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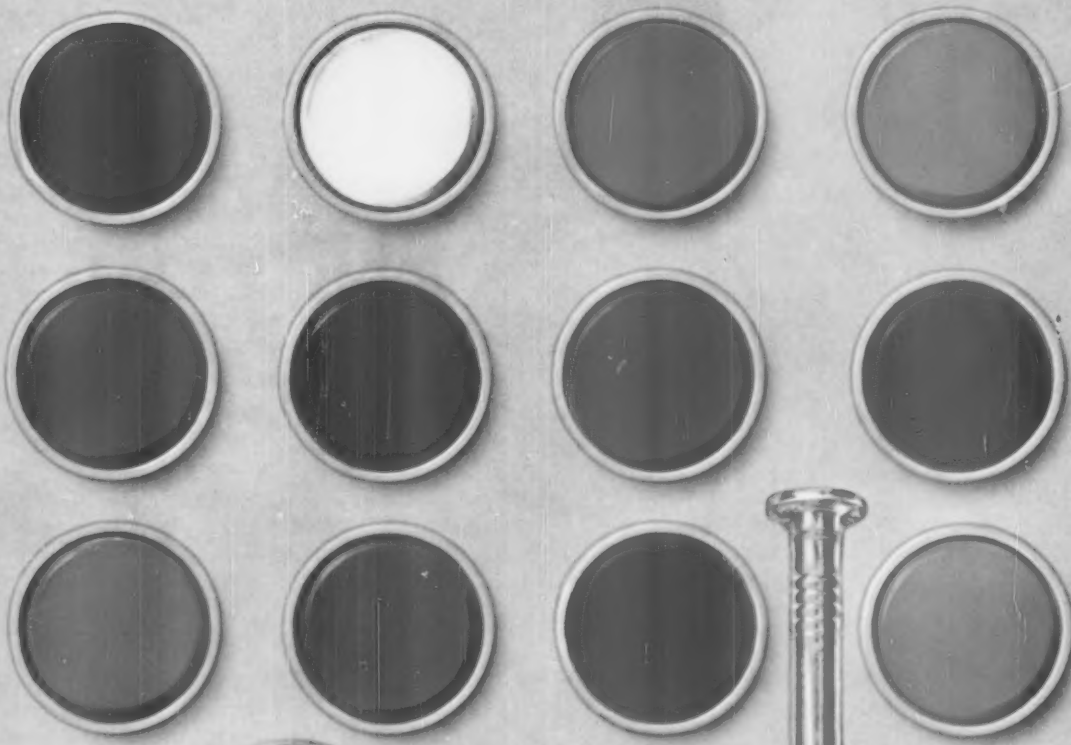

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Activities of Annual Meeting Committee

Pictured on this page are the busy IFT'ers who have been diligently applying themselves for six months and more in planning the 1961 Annual Meeting in New York.

General Arrangements Chairman E. Paul Orsay has been working closely with previous chairmen and IFT Headquarters to determine areas of shortcoming in the past. His idea is to profit from previous errors by applying appropriate ideas and suggestions gleaned over the years, to this meeting. Mr. Orsay is responsible for the smooth functioning of the entire five-day Meeting, which includes the operation of the following committees:

E. P. Orsay

Program. Mr. Fred Baselt has completed much of his work as chairman. One hundred sixty-nine papers are scheduled, including five symposia. Sessions will include: Microbiology, Enzymes, Flavors, Dehydration, Fruits and Vegetables, Pressurized Food Products, Packaging Food Products, Meat and Fish, Colors and Flavors, Pigments and Lipids in Animal Products, Statistical Techniques, Process Engineering-Instrumentation, and General Research. Featured symposium will be "Food Science and Feeding Mankind," on Tuesday morning. NO CONCURRENT SESSIONS ARE PLANNED while this is in progress. Other symposia are: "Role of Food Technology in Civil Defense," "Food Additives," "Statistical Quality Control," and "Education of a Food Technologist."

Sessions and Banquet. All organized meal planning, including the Awards Banquet, comes within the realm of Chairmen E. E. Macdonough and P. X. Hoynak. This major area of concern is assignment of rooms for technical sessions, including projection equipment, operators, public address systems, and proper door signs, for your convenience, as well as meal functions.

F. Baselt

P. X. Hoynak

E. E. Macdonough

Field Trips. Dr. J. C. Bauernfeind has completed arrangements for eight field trips, including two all-day trips and six half-day tours. Further details are printed elsewhere in this issue.

General Entertainment. Assuming that our appetites for knowledge will be somewhat satiated after the daily technical sessions, Dr. K. T. Farrell, Chairman, plans rest and relaxation for all. Tuesday, this will be in the form of the Awards Banquet, which will feature an excellent meal plus entertainment of a top-flight nature.

Ladies' Entertainment. Miss Terry Pavlik has arranged three tours to: General Foods Kitchens, Radio City, plus a cruise, each designed to give visitors a rapid yet broad look at the New York area. As chairman she has also planned the Sunday afternoon Welcome Tea, but these items are spelled out in the special invitation letter, included with your registration and ticket request envelope.

J. C. Bauernfeind

K. T. Farrell

T. Pavlik

Courtesy. Dr. G. E. Livingston hopes to make your stay in New York more comfortable, by providing an information service which will cover everything from garage locations to a service which will aid in finding "lost" meeting rooms, etc.

Publicity. To aid in keeping the public aware of the significance of this Meeting, Mr. Tom Gartland will maintain liaison with local and national radio, television and press services. Since some papers may merit national recognition, he will ensure that they are given due credit.

Employment. Mr. W. X. Clark had graciously accepted chairmanship of this committee; however, the IFT Employment Committee recommended, subsequent to his acceptance, that an individual not connected with a food industry firm be named chairman. Mr. Clark agreed to this proposition, and Mr. Claude Cruse, Production Manager of FOOD TECHNOLOGY, has been named Chairman. New operating policies, designed to prevent any compromising situations from arising, are described in your invitation to the Meeting.

G. E. Livingston

T. Gartland

C. J. Cruse

Registration. Mr. H. Chenoweth, Chairman, will be responsible for proper manipulations for the few individuals who have not taken the opportunity to pre-register. It is hoped that everyone planning to attend the Meeting will also submit their registration form and covering check (where applicable) in accordance with instructions in your invitation letter.

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MARCH COVER. Termed a "Valley of Fermenters" by Miles Chemical Company, the row of citric acid fermenters depicted is used by that company in the manufacture of citric acid. Rather than utilizing the more familiar sources of citric acid production, such as the citrus fruits, this process involves a deep fermentation process. A mutant of *Aspergillus niger* converts sugar into citric acid; this acid is said to be of the highest purity and quality. Adaptation of this process to large scale production has made it possible to produce great quantities of citric acid for the open market. In addition to uses within the food industry, citric acid is used in the pharmaceutical field in certain medications, and in cosmetics such as skin lotions and hair rinses. Industrial uses include detergents, rust and scale removal, secondary oil recovery, electroplating, tanning, printing, ion exchange, and sea water evaporators.

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Impact of the New Food Law Amendment on Research in Food Technology*

Margaret Ives

Senior Research Toxicologist, Research Center, American Can Company, Barrington, Illinois

THERE HAS BEEN a lot of speculation since September, 1958, about stifled research in food technology because of the unreasonable requirements and time limits of the new food-law amendment. Many remarks quoted in the press deplored the plight of the housewife: "she will no longer be able to buy convenience foods, particularly the unusual or gourmet type"; "she will see no future advances in food packaging"; "the price of processed foods will increase"; etc.

Well, two years have elapsed and as far as we can see none of these dire predictions have come to pass. This is usual. Man always resists changes, but shows a happy facility to adjust to them rapidly when such changes occur. Some benefit usually accrues from any change in policy, although many problems may also be generated.

The fundamental problems generated by the administration of this law to new products are twofold: increased cost of development, and a long interval between developmental work and commercial use because of long-term chronic testing on animals. Because this interval may be as high as seven years and the costs may easily exceed $250,000, it follows that competition may decrease since there will probably be fewer new products on the market.

There are diverse opinions among scientists as to the best solutions of these problems. The suggestions I am making reflect my professional opinion as a toxicologist, but do not necessarily reflect the opinions of the Institute of Food Technologists or of my employer.

The immediate answer to the problem seems to be increased basic biological research, perhaps at the cellular level, where rapid growth might shorten the time required to detect toxicants. This is the approach that many technical people feel should be used. The study will take several years; thus we are back to the beginning, with prohibitive costs. Some of the companies with large research laboratories will take the applied research approach to keep their products on the market and devote the major proportion of their efforts to chronic-feeding studies. This leaves practically no time or money to study new approaches to biological investigations. It appears that academic institutions will have to fill the gap.

In addition to the long observation periods required to comply with the regulations to prove safety, emphasis is also placed on the presence of trace amounts of materials in foods. Elaborate analytical techniques are often required to determine the presence of such materials in food at the level of one part per million or less. Development of these techniques is time-consuming and requires a high degree of professional skill that most companies cannot assign exclusively to food-additives problems.

ENLARGEMENT OF RESEARCH PROGRAMS IN FOOD TECHNOLOGY

We have delineated the problems. Now, what is being done about them from a research angle? The academic institutions are enlarging their departments of food technology or pooling specialists from various departments to conduct fundamental biological research on the physiological effects of low-level chronic feeding. Various types of materials will be studied. The University of California has established on the Davis Campus a Pesticide Residue Research Section to study the biological effects of pesticide residues on warm-blooded animals and to develop new analytical techniques for measuring such residues. In addition, plans have been drawn up for a group to study the effects of food additives on single-cell organisms such as bacteria, yeasts, and molds and on embryonic tissue. This is an attempt to decrease the time required for chronic-feeding studies. Of course, any investigation of that nature will be time-consuming since results will have to be correlated with long-term animal studies before reliance can be placed in the methods.

Purdue has established a Trace Level Research Institute for residues and metabolites. The membership of this group is drawn from many departments, and the facilities will be made available to industrial interests such as food and fiber producers, and to the Government. The Institute plans to do basic research in pharmacology, toxicology, metabolism, and residue problems. A similar institute exists at Syracuse University to aid the paper industry, while the Pharmacology and Toxicology Departments at the Universities of Rochester and Miami, which have engaged in this type of research for years, have widened the scope of their activities.

Stanford University has received a grant of $300,000 to study the effects of pesticide residues on man. It is their hope to develop a method for the rapid diagnosis of chronic pesticide poisoning in man. An earlier grant, which was also from the John A. Hartford Foundation, resulted in the development of an automatic analyzer for pesticide residues. The

* Presented before the Joint Meeting of the Food Law Institute and the Food and Drug Administration, November 28, 1960, Washington, D. C.

Massachusetts Institute of Technology also has plans for extending food technology research into the area of food additives. With this mention of only a few of our academic institutions that are planning to study this problem, it is apparent that they are stepping into the gap mentioned earlier.

Industry has increased its emphasis on this type of research to some degree, even though it is applied rather than fundamental research. Many suppliers of basic raw materials, who have never been concerned with the uses to which their products were put, have found themselves questioned about biological activity and migration. In some fields, such as steel, aluminum, resins, inks, plastic, adhesives, and paper, many of the organizations affected had no one qualified to answer such questions. Several companies established animal laboratories and hired toxicologists to study their individual problems; others hired toxicologists as consultants, and biological testing of their materials was carried on at outside consulting laboratories. These particular manufacturers would probably not have become involved in this type of work had it not been for pressure from their customers, since they did not know all the uses of their products and the significance of these uses to public health.

Food producers and packers panicked at the wording of the new amendment and demanded unconditional guarantees of safety from suppliers. Actually, a good many of these warranties are worthless since the customer changes the material in some way after purchase in order to manufacture his product; hence, the original supplier cannot be expected to be responsible for the altered product.

One absurd example of this situation was reported in May, 1960. A chain store required a food processor to supply it with guarantees that their food product complied with the new food law amendment. The processor decided not to take the full responsibility; he contacted the grower and demanded a guarantee from him. The grower, remembering the cranberry episode, decided to close all loop-holes and demanded such a guarantee from the fertilizer salesman. The reply was "Fertilizers are not for human consumption, they go in the ground."

Not only has biological research been stimulated in the industrial laboratories of raw-material suppliers, but also the size and number of biological consulting laboratories has multiplied many times. This reflects the amount of money being spent outside of the industry's own laboratories to solve these problems.

Recently, more research in government laboratories as well as government-sponsored research in academic laboratories has been directed toward the food-additives problem. United States Department of Agriculture laboratories, especially the Western Regional Laboratory, have been quite active. Scientists in the Food and Drug Administration have also been active in the field of basic research, although their time has

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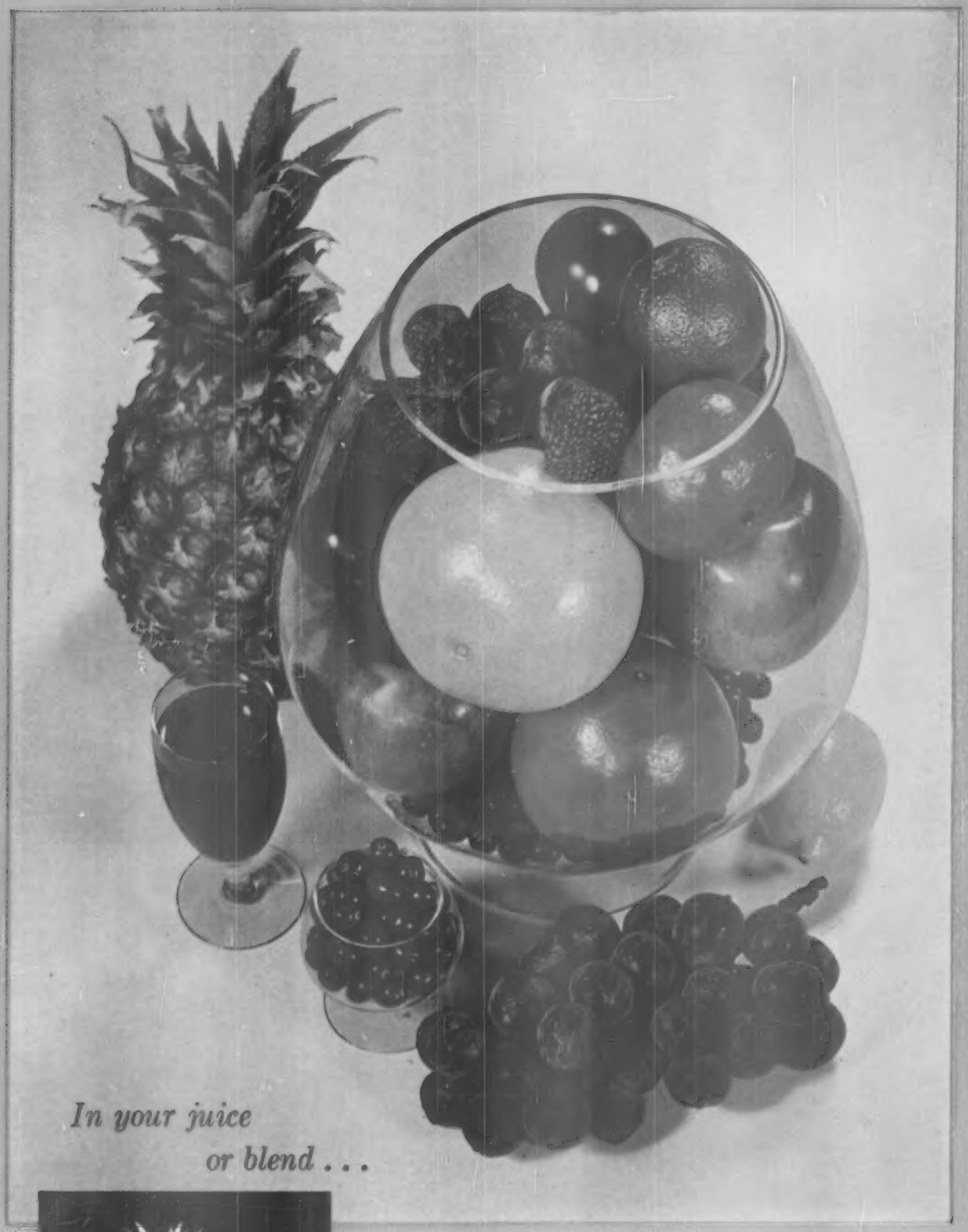

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been limited by the great pressure of procedural and administrative work generated by the law.

INCREASED EMPHASIS ON ANALYTICAL TECHNIQUES

Experts in two scientific disciplines are required to devise and apply testing procedures under the regulations of this law. The first is the toxicologist, whose biological research we have been discussing, and the second is the analytical chemist. After the biological testing period is over and the material is considered a direct "food additive," a suggested tolerance with accompanying analytical test will be required in the formal petition. The analytical chemist must develop this test. The gas chromatograph and the spectrophotometer are instruments frequently required for these analytical procedures.

In addition to direct food additives, there is also the problem of extraction from the container, and hence, indirect food additives. The problems of extractives and pesticide residues are similar because they involve extremely small amounts of a substance and very sensitive analytical procedures. A short course on extraction procedures was held in March, 1960, at Michigan State University in cooperation with the Beckman Instrument Company. This workshop was particularly valuable to those analysts, especially from industry, who were not familiar with the ramifications of the law.

Analytical laboratories in both universities and industrial concerns have acquired these newer instruments. Not only have they been used to determine residues of pesticides, antibiotics, and migratory materials, but they have also been applied to other problems in food technology. Considerable progress has been made in the study of the constituents of natural products such as the volatile components of coffee, lemon oil, and vegetable flavors such as peas and cabbage. Knowledge of the composition of natural products permits the manufacture of synthetic materials with the same general properties. Often, only in this way can a product be freed of a toxic natural constituent. Decaffeinated coffee is an example of such a product.

Even though the faddists would have us believe to the contrary, naturally occurring products are not, *ipso facto*, safe, or even wholesome and nutritious. Rhubarb, with its extremely high level of oxalic acid, is an example of a natural product that is not completely safe. Oxalic acid is present in the leaves at toxic levels; the more moderate level in the stalk is present in an amount that reacts with the available calcium in the diet to form an insoluble oxalate so that the calcium is unavailable to the body. Such an unaltered product is not covered by the food-law amendment.

INCREASED EMPHASIS ON FUNDAMENTAL BIOLOGICAL RESEARCH

Another positive gain from research, related to the food-additive area, lies in the field of metabolic

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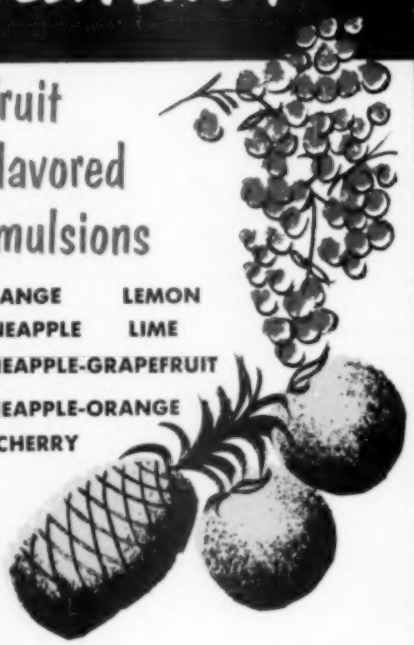
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studies. A knowledge of the site of detoxification, metabolic end products, and amount of actual tissue absorption is necessary for assessing completely the toxicity of any material. In a metabolic study one frequently learns that the toxic material is not the original ingested substance but a metabolic end product. Bonzer and co-workers, in England, demonstrated this in the case of 2-naphthylamine.

If such a situation develops it may be chemically possible to alter the original material so that it is metabolized by a different pathway while the desirable chemical properties are maintained. Information gained in these studies about metabolic pathways and enzyme activity is useful in other biochemical and nutritional studies.

The biochemical and pathological lesions developed in a metabolism test are clues to the outcome of a chronic test. In certain instances, but unfortunately too seldom, such a wealth of information has been acquired from the metabolic study that a long-term chronic study has not been required to clear the material.

UNDUE EMPHASIS ON TRACE CONTAMINANTS

Some positive and negative aspects of the influence of this law on research in food technology have been cited. Now, what is the net result of these aspects on research in general? For one thing, undue emphasis has been placed on trace contaminants migrating from a packaging material, and on pesticide and antibiotic residues of less than one part per million.

There seems to be an advantage in using certain antibiotics that are not of therapeutic value in the preservation of food. These particular antibiotics will not cause the development of resistant strains of pathogenic bacteria. They aid in the preservation of meat in areas with poor refrigeration; they decrease the time and temperature of the heat process for canning; they are useful as a dip in preventing surface contamination of frozen meat and poultry. A test has been developed for determining the presence of an antibiotic at a level of 0.005 ppm in animal tissue. Will tolerances under the food-additive law be set at such levels? Are we not straining at gnats? Also, this particular procedure points up the fallacy of prescribing a tolerance with a "practical zero," which implies zero residue when analyzed by a specific method. Such a tolerance acts almost like a challenge to the development of more sensitive methods.

UNDUE CONCERN ABOUT DIETARY CARCINOGENS

Another negative aspect of this law is the undue concern about dietary carcinogenic activity of materials of unknown biological activity at low levels. Attempts to prove a negative are time-consuming and expensive, and only meager scientific information is obtained as a result of the necessary statistical design. A case in point is the American Petroleum Institute's program to prove the safety of the paraffin waxes used in contact with food in America for several generations. The study has continued for several years; it

has involved a great amount of animal experimentation and the services of several highly skilled scientists. Could not this money and effort have served Man better if applied to the study of the causes and cure of cancer?

EFFECT ON NEW DEVELOPMENTS

Yet a third negative aspect lies in the applied research field. In the spring of 1960, one company announced that it was stopping research in the field of medicated animal feeds because the work involved in determining the minute residue in the tissues and the effects of this residue on other animals and Man was too time-consuming and expensive. Certainly, we know if a company cannot make a profit on an item, it cannot manufacture it.

New adjuncts, flavors, colors, and packages for foods have been developed continuously by the leaders in the field. Will this rate of development continue? We hear many say "no," because they cannot afford the testing time and cost. The margin of profit in items in the food field is low. We cannot have the true answer to this question for a year or more, since many of the apparent new products today were actually in commercial use in certain areas prior to January, 1958. The question really is: shall we have as many new food products and advances in packaging in January, 1963, as we had in January, 1959?

Dr. R. L. Hall, of McCormick and Company, states, "Almost certainly in the flavor field a totally new food additive could not possibly justify the risks and expenses involved in assembling the information required in a petition. We must rely largely upon those ingredients with a history of use whether they are used intentionally as such or occur naturally in foods."

Dr. Adolph Miller, of Milprint, Inc., predicted in a talk before the Packaging Institute's recent national seminar: "Packaging costs will rise due to increased developmental costs and broadened financial responsibility given suppliers in the form of guarantees. These added costs will be reflected in rising prices of finished products to consumers. It is obvious that the people, as represented by Congress, are prepared to accept the new costs, since the security obtained by the legislation is considered desirable."

NET RESULT OF IMPACT OF NEW FOOD LAW

The three negative aspects cited can be set against three positive ones, also discussed earlier. These may be stated in a few words, but the impact and knowledge gained to help Man are far weightier.

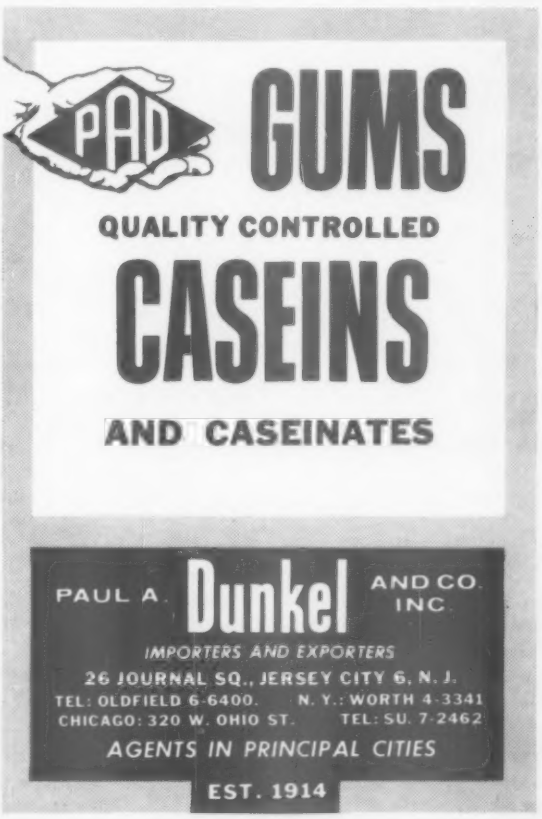
The expanded academic research as planned at California, Purdue, Stanford, and many other strong institutions, will increase basic information about the metabolism of foreign substances in the animal body. These studies may shed light on biological phenomena and may advance medical research.

Being developed in industrial, governmental, and academic laboratories are new analytical tools that may aid in many fields, from determining the purity of water and the atmosphere, to the study of archeological relics.

The third positive aspect lies in the migration studies. Industrial polymer chemists are extensively investigating coatings, adhesives, and other materials to determine what might migrate into foods from containers. The identification of these migrants sheds light on what compounds may be formed during the curing cycles. Further improvements of materials may result from these required tests; who knows?

All in all, the negative aspects of this law appear to be related to human fallibility, whereas the positive aspects are concerned with "pushing back the frontiers of science," to quote the motto of the Gordon Research Conferences. Let us cease to be concerned with striving to perfect precise measurements of less than one part per million. Let us keep known dietary carcinogens out of our food and investigate any suspicious new materials, but not spend a fortune trying to prove a negative with an innocuous material used at extremely low levels. Let us be realistic in our judgment of food additives and not prevent the marketing of materials by demanding unnecessary and expensive testing.

If we do not strive for safety proved to infinitesimal levels, but consider all the practical solutions to the problems, the new food-law amendment will more than justify its existence by protecting the public health and encouraging advancement in knowledge.



Council Actions—Midyear Meeting

Results of the Council Ballot on which recommendations were made by the Executive Committee at the Mid-Year Meeting, show that all items were approved by a significant majority.

NEW EXECUTIVE SECRETARY

Mr. Ray B. Wakefield, Chairman of the Committee on Administrative Policy, announced the appointment of Mr. Calvert L. Willey as Executive Secretary of the Institute. He will replace Col. Lawrence, who will retire June 30, 1961. Mr. Willey will come with the Institute on May 1st, and will actively assume the duties of Executive Secretary at the close of the New York Annual Meeting. Our new Executive Secretary is currently holding a similar position with the American Society of Lubrication Engineers (ASLE) in Chicago.

C. L. Willey

Mr. Willey was born in Maryland and attended schools there, culminating with a B.S. degree in Business Administration from the University of Maryland in 1949. Prior to his association with the ASLE in 1957, he was Assistant to the Executive Director of the National Society of Professional Engineers, and from 1952 to 1954 he was a research assistant with the Clifton Corporation. Mr. Willey is currently a Lieutenant in the Naval Reserve, and served on active duty between 1942 and 1952.

NEW IFT JOURNAL POLICIES

A new statement of policies relating to publication of IFT Journals was approved. Portions are quoted in order that the membership may anticipate forthcoming changes, and understand the new Editor's justifications for these changes.

Food Technology

"*Mission.* The mission of Food Technology will be to promote the professional interests of scientists, engineers, and technologists concerned with manufacture, processing, storage, transport and marketing of foods. A special effort will be made to serve the specific interests of members of IFT. This mission will be accomplished by publishing: (1) feature articles on trends in food processing and marketing, (2) special articles on current developments in closely related fields, such as crops and livestock production, human nutrition and public health, (3) research papers on food engineering, new product and process development, storage of foods and other technological subjects, (4) editorials and commentaries on subjects of professional interest to food technologists (education, organization and management, promotion of food technology as a profession, etc.), (5) news and notes concerning IFT activities as well as those of related technical societies, and (6) reviews and abstracts of current technical literature relating to foods and human nutrition. A Board of Editors, consisting of ten members (including the Executive Editor as chairman), will be responsible for review and approval of research papers submitted for publication in Food Technology. The Executive Editor and members of the Board will name several panels of qualified reviewers, each of which will represent a certain subject matter area.

"Review of research papers will be carried out as follows: Manuscripts will be submitted to the Editorial Office by the authors. The Executive Editor will first determine whether or not papers are generally suitable for publication in Food Technology. If not, he will return them to the authors with an appropriate explanation. If suitable, he will submit them to qualified members for editorial comment and recommendation. Board members may review articles; however, ordinarily they will be forwarded to a member of one of the review panels. The Executive Editor has the sole responsibility for the final acceptance or rejection of manuscripts."

Food Research

"*Introductory Statement.* There has been a good deal of dissatisfaction with the type and quality of articles published in Food Research. Critical review and appraisal of this problem suggests that fairly drastic steps are needed to guarantee that this research journal be of the highest quality in the future.

"*Mission.* The mission of the publication will be to promote the interests of research in food science. The mission will be accomplished by publishing research articles of the highest quality in this field. Articles will be published on subjects such as the following: Physical and chemical studies on the separation, isolation, identification, characterization and reactions of components of foods; chemical composition and nutritive value of foods; microbiology of foods; toxicology and nutrition; and the sensory evaluation of foods.

"*Name of Journal.* The name of this journal will be changed to Journal of Food Science. For a limited period the cover and title page will carry the statement "Formerly Food Research." The numbering of the volumes will continue as for Food Research.

"A Board of Editors will be named and will consist of twelve members; eight of these will be from the United States and Canada and four from other countries. (The latter to be called International Editors.) The Executive Editor will be ex-officio chairman of the Board. The Executive Editor and other members of the Board will name panels of qualified reviewers, each of which will represent a certain subject matter

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area. Review will be handled as follows: Manuscripts will be submitted to the Executive Editor or to one of the International Editors. A decision will first be made as to whether or not the manuscript is generally suitable for publication in the JOURNAL OF FOOD SCIENCE. If not, it will be returned to the author with an appropriate explanation. If suitable, it will be submitted to a qualified panel member for editorial review and comment. The Executive Editor will have the sole responsibility for acceptance or rejection of papers submitted for publication in this journal.

"The JOURNAL OF FOOD SCIENCE will carry only a nominal amount of advertising. All advertising copy will require specific approval by the Executive Editor."

In keeping with the newly approved policy that the Editor of Institute Journals be given full responsibility for appointment of Editorial Boards as to name, number, and term of office, your Executive Editor has named the following to membership on these Boards: FOOD TECHNOLOGY—C. P. Bolton, Ben F. Buchanan, Richard H. Forsythe, J. M. Jackson, Maynard A. Joslyn, Reid T. Milner, A. I. Morgan, Jr., James W. Pence, and W. M. Urbain; JOURNAL OF FOOD SCIENCE (United States and Canada)—Jean F. Caul, Robert E. Feeney, Calvin Golumbic, Z. I. Kertesz, Leo Kline, C. F. Schmidt, and H. L. A. Tarr; (International)—J. Kuprianoff, Tetsujiro Obara, J. R. Vickery, and one additional person to be named.

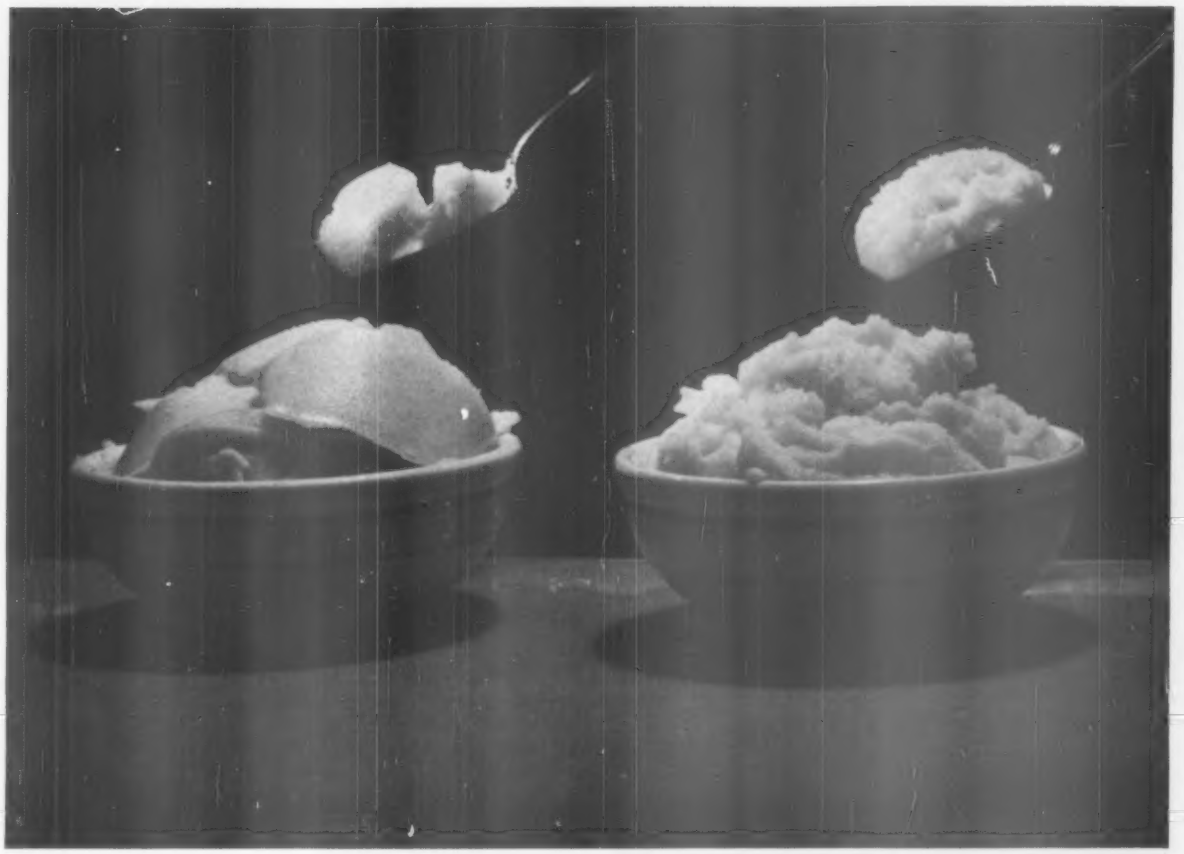
Council has also given its consent to the proposal that the name of FOOD RESEARCH be changed to JOURNAL OF FOOD SCIENCE, beginning January, 1961.

Another proposal advanced was that the IFT turn over the JOURNAL OF FOOD SCIENCE to the Academic Press (New York City) with the exception of copyright ownership. This was referred to the Publications Committee for further study, with a request for a report at the next meeting.

A standing IFT committee will be appointed in the near future to maintain liaison with the FDA. Although duties are not yet strictly defined, approval was given to the idea of setting up joint IFT-FDA forums to exchange views on technical problems. The committee may also be concerned with encouraging the FDA to publish in FOOD TECHNOLOGY, articles which would better inform our members about FDA, its problems, and its need for technical information and assistance. To aid further in keeping members informed, IFT Headquarters will apprise the various Regional Sections of FDA officials who are available as speakers. In turn, we shall, upon request, provide the FDA with names of experts in certain areas. Under this policy, it is possible that Members could be called upon to render their services in a professional capacity to the FDA. Such service would be on a voluntary basis.

REPORTS

Treasurer Dr. Prater stated that the Institute is still in good financial condition. Current yield in the Investment Account is 3.5%, and an additional trans-



Try this texture trick

Pour ½ cup of plain, dry farina and a teaspoonful of salt into 3 cups of boiling water and cook until done. To another ½ cup of the same dry farina, add 3% of *Myverol® Distilled Monoglycerides, Type 18-07*, little white beads closely resembling the farina itself in appearance. (Write us for a free sample.) Pour *that* with its teaspoonful of salt into 3 cups of water, not boiling but at any temperature below 140 F. Then boil *that* until done.

Now set both pans aside for a few hours. Keep them both warm or let them both cool. It doesn't matter.

Come back and note which panful has turned into a jellylike mass and which still looks and behaves as it did at the moment the cooking was finished. See if you can taste a difference. (You can't.)

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fer of $25,000 to $50,000 to this account will be effected.

In other financial matters, approval was given to the Editor to expend $846.00 on a FOOD TECHNOLOGY Readership Questionnaire. To cover expenses above and beyond the appropriation approved last May providing for editorial overlap costs, $3,200 has been appropriated for Editorial Office Maintenance and Travel. $1,000 was approved for moving IFT Headquarters to new offices (Suite 1350) in the same building, where about 50% more space is available. $100 each were voted for legal fees and payroll taxes.

OTHER HIGHLIGHTS

The Executive Secretary reports that as a result of a mailing, the IFT has increased its membership by over 200, and it is anticipated that we will end the fiscal year with over 6,000 members.

The following items were also approved:

1) The IFT has retained Winston, Strawn, Smith & Patterson as legal counsel, and their name is on all official stationery.

2) Each Past President shall receive an appropriate plaque at the meeting immediately following the expiration of his term of office as President.

3) The April issue of FOOD TECHNOLOGY shall carry a directory of Exhibitors at the 1961 Annual Meeting.

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International Food Technology

Australians are making comprehensive plans for the 12th World's Poultry Congress to be held in Sydney in August, 1962, with American participation an important part of those plans.

So reports **Dr. Cliff D. Carpenter,** chairman of the U. S. Participation Committee, following a meeting with J. E. Beveridge, head of the Industrial Public Relations Service of Sydney.

Beveridge recently visited the U. S., and contacted individuals and companies regarding their participation in the Sydney Trade Fair in 1961 and the World's Poultry Congress the following year. Accompanied by his wife and daughter, Beveridge traveled more than 37,000 miles on his assigned missions, visiting 26 countries in about five months.

At Chicago he met with Dr. A. W. Jasper, secretary of the U. S. Committee, and Lee Campbell, chairman of the U. S. Committee's subcommittee on exhibits. In San Francisco, Beveridge was met by Vice-Chairmen **Dr. Hans Lineweaver** and **Dr. A. W. Brant,** and Secretary Jasper.

Returning from a trip to England and Western Europe, Dr. and Mrs. Carpenter met the Beveridges at Los Angeles just prior to their departure for Honolulu, the last leg of their return journey to Australia.

Beveridge reports enthusiastic reception around the world for the Poultry Congress. He was well pleased with the progress made in the U. S. on organization and development plans.

Australia's Prime Minister, the Right Honorable R. G. Menzies, has accepted the position of Honorary President for the 12th Congress. Seventy-four countries have received official invitations from the Australian government, to participate in the Congress.

Pre-Doctoral Research Fellowships Available

Several pre-doctoral research fellowships and assistantships in the Institute of Nutrition and Food Technology are still available at The Ohio State University.

The graduate program pursued will depend upon the interest of the student, and will lead to the Ph.D. degree in a University department representing that interest. Some of the departments concerned are: Agricultural Biochemistry, Animal Science, Bacteriology, Dairy Technology, Home Economics, Horticulture and Forestry, Physiological Chemistry, and Poultry Science.

Specific areas in which research is being done are rumen metabolism, embryo nutrition, intermediary metabolism, human nutrition, factors in food spoilage and flavor changes, studies in toxic residues in food, food microbiology, and the biochemistry of milk, meats, and endocrine functions.

Research under the tenure of these fellowships and assistantships will normally be carried out in the Institute of Nutrition and Food Technology Laboratory,

The Influence of Sugar in Ice Cream. III.

Effect on Physical Properties

T. A. Nickerson and Rose Marie Pangborn

Food Science and Technology, University of California, Davis, California

(Manuscript received October, 1960)

SUMMARY

A study was made of the effect of sugar on some of the physical properties of ice cream. Its effect on viscosity, melting rate, and firmness was evaluated. The influence of other compositional changes was also determined.

Increases in viscosity were only small from increasing the sugar content from 13 to 19% or the milk solids-not-fat from 10 to 12%. Stabilizers, causing a marked increase, had more effect on viscosity than any other ingredient. Melting rate was not appreciably affected by the differences in composition studied. Firmness, determined by penetrometer measurements, was affected most by storage temperature. Increasing sugar content softened the product noticeably. So did increasing the overrun. Stabilizers tended to maintain a more uniform firmness with changes in overrun. Seeding the ice cream to crystallize the lactose produced a firmer ice cream.

RECENT INVESTIGATIONS (Pangborn and Nickerson, 1959; Pangborn *et al.*, 1957) have shown that consumers prefer relatively high sugar levels of 17–19%. The ice cream industry, however, has accepted 15% sugar or its sweetness equivalent in a combination of sucrose and corn syrup. The industry has been reluctant to adopt higher sweetness levels, because physical properties might thereby be changed.

Sugar, besides being essential for palatability, is important to the physical properties of ice cream and its unfrozen mix. Sugar influences body, texture, firmness, freezing properties, and melting characteristics. Any changes in these properties due to changed sweetness level would be of prime interest to an ice cream formulator.

This study concerns some of the factors that control viscosity, firmness, and melting characteristics.

EXPERIMENTAL METHODS

Ice cream samples. Ice cream was processed in a manner normal in commercial operations. All samples contained 12% butterfat, but the other constituents—milk solids-not-fat, sugar, and stabilizer—were varied. The mixes were pasteurized 30 min at 160°F, homogenized at 2,500 and 500 psi, cooled to 40°F, and stored overnight before freezing in a continuous freezer. The ice cream was quickly hardened and stored in cabinets at −10°, 2°, and 12°F until tested.

Viscosity. Viscosity measurements were made on a direct-reading (Fisher) electroviscosometer using the small (XI) bobbin. Determinations were at 40°F.

Melting characteristics. A cylinder of ice cream 1½ in. high and 3¼ in. in diameter (top 1½ in. from a pint Sealright container) was placed on ¼-in. wire screen above a funnel. Melting rate was measured by the volume of drip from the funnel into a graduate cylinder. The volume was recorded at 5-min. intervals during a 30-min. test period.

Firmness. Firmness was measured by a cone penetrometer. The procedure and equipment were similar to those used by Haighton (1959) to measure the hardness of fat and margarine. A metal cone of 40° angle and total weight (cone plus moving parts) of 178 g was allowed to penetrate the sample for 5 seconds. Firmness was expressed in terms of penetration depth, measured to the nearest 0.1 mm.

RESULTS AND DISCUSSION

Effect of sugar. The data in Table 1 show that varying sugar content between 13 and 19% had little effect on the viscosity of the mix. As the sugar was increased the viscosity also increased, as expected. However, the increased viscosity was greater than would result from a similar addition of sugar to a sugar solution. These results are typical of those obtained throughout the study. The viscosities in Table 1 are lower than usual. Other mixes of similar composition, but processed at other times with different milk products, had considerably higher viscosities. Differences in chemical composition and processing treatment are known to have a marked influence on the viscosity of milk (Sommer, 1951).

Table 1. Effect of sugar on viscosity and melting rate.[a]

	Sugar levels			
	13%	15%	17%	19%
	(cp)			
Viscosity of mix	25.4	27.8	31.0	34.4
Melting rate[b]	(ml)			
Time (min)				
5	0	0	0	0
10	0	0	0	0
15	0	0	0	0
20	2	3.5	5	4
25	8	9	14	13
30	15.5	18	23	24

[a] Composition: 12% fat, 11% milk solids-not-fat, no stabilizer.
[b] Ice cream at 2°F, rm. temp. 72°F.

With increase in sugar content, there was a slight increase in melting rate. The differences were of low magnitude, however, and insignificant from a practical standpoint.

Effect of milk solids-not-fat. Milk solids-not-fat in ice cream are normally 10–12%. The data in Table 2 show the influence of variation within this range on some of the physical properties of the mix and ice cream.

The samples represented there differ from those in Table 1 by containing 0.35% commercial stabilizer (Hi Gel), with much higher viscosities resulting. The increase in viscosity due to greater milk solids-not-fat is small compared to that from increasing sugar content.

These data also show that the melting rate did not vary greatly with the compositional changes used

Table 2. Effect of sugar and milk solids-not-fat on viscosity, melting rate, and firmness [a]

	Sugar and milk solids-not-fat levels					
	13% sugar 11% MSNF	15% sugar 11% MSNF	17% sugar 11% MSNF	19% sugar 11% MSNF	15% sugar 10% MSNF	15% sugar 12% MSNF
	(cp)					
Viscosity	136	149	160	230	143	172
	(ml)					
Melting rate [b] Time (min)						
5	0	0	0	0	0	0.5
10	0	2	1.5	2	2	2.5
15	2.5	9	9	9	8	9
20	10	18.5	21	20	20.5	18.5
25	20.5	31	36	32	33	31
30	31.5	41	49	47	45	43
Firmness (penetration depth)	(mm)					
2°F	12.4	14.4	17.2	17.8	13.1	14.8
12°F	23.2	27.0	28.0	37.9	26.2	27.4

[a] Composition: 12% fat, 0.35% Hi Gel stabilizer.
[b] Ice cream at 12°F, rm. temp. 78°F.

here. Firmness was little related to melting rate. On the other hand, differences in firmness due to differences in composition are shown by the penetrometer data. Softening due to increased storage temperature is also apparent from the penetrometer data. Differences in firmness due to differences in storage temperature were much more pronounced than the effects of any compositional change.

Effect of overrun. Another compositional variable in the production of ice cream that can influence its physical properties is the overrun (percentage of air incorporated during freezing). There was a straight-line relationship between overrun and firmness as measured by the penetrometer when no stabilizer was used in the ice cream (Fig. 1). Stabilizer tends to maintain a more uniform firmness over the range of overrun normally encountered in commercial ice cream.

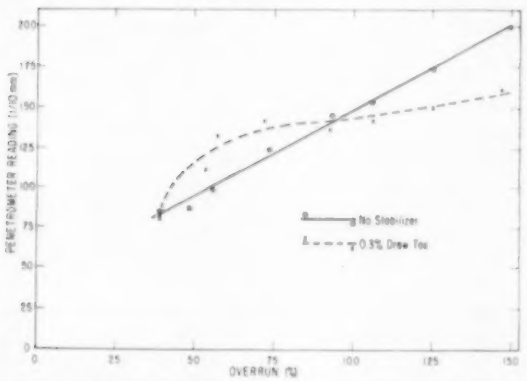

Fig. 1. The effect of overrun on the firmness of ice cream with and without stabilizer.

Effect of seeding (lactose crystallization). Seeding of ice cream (Nickerson, 1954) to force rapid crystallization of the lactose has been reported to produce a drier, firmer ice cream (Decker, 1956). Increased firmness from seeding is apparent from the data in Table 3. The lactose crystallizes rapidly on the nuclei produced by the seeding (Nickerson, 1956). The freezing point rises as the lactose is removed from solution, and freezing is therefore more complete. This is another means of producing a firmer product.

Table 3. Firmness of regular and seeded ice cream.

Composition of ice cream [a]	Penetration depth at 2°F (mm) Not seeded	Seeded
1. 11% MSNF	13.6	9.0
2. 12% MSNF	12.0	9.0
3. 12% MSNF	11.2	6.3
4. 12% MSNF, 0.3% Hi Gel	11.7	9.0
5. 14% MSNF	15.4	9.5
6. 14% MSNF	10.3	8.4
7. 14% MSNF, 0.3% Dariloid	13.6	6.9
8. 14% MSNF, 0.5% Hi Gel	11.3	7.2
9. 14% MSNF, 0.3% Dariloid, 0.2% Atmul	10.9	7.2

[a] All contained 12% fat and 15% sugar.

Acknowledgment

This study was supported in part by funds from the Sugar Research Foundation, Inc., New York.

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Factors Affecting the Development of "*Bacillus coagulans*" in Fresh Tomatoes and in Canned Tomato Juice[a]

A. H. Jones[b] and W. E. Ferguson
Food Processing and Storage Plant
Research Institute C.E.F., Ottawa

(Manuscript received August 12, 1960)

SUMMARY

One of the problems encountered in studies on *Bacillus coagulans*, the organism associated with "flat-sour" tomato juice, has been the difficulty experienced in growing the test organism in the usual canned juice. Studies with both the vegetative and spore stages of this organism have indicated that tomato juice generally is not a favorable medium. Growth will develop only under specific conditions and particularly when the pH of the juice is altered to the less acid state. In view of the importance of pH in the development of *B. coagulans*, studies have been conducted to determine conditions conducive to the development of this organism in tomato juice.

Results of studies have indicated that tomatoes held in the field after harvesting, a common practice used to develop a better color, or held at the plant before processing, invariably become less acid and pH readings as high as 5.00 have been recorded on individual tomatoes. The change in pH will depend to a considerable extent on the temperature of holding; the higher the temperature within a range of 20° C to 45° C, the greater the change.

During the past season it was found that tomatoes sprayed with a suspension of *B. coagulans* at various stages of the holding period showed a marked increase in the rate of growth of the test organism coinciding with the change in pH. This increased rate of growth occurred when the pH reached 4.30 and continued until the appearance of heavy mold contamination. At this time both increases and decreases in the rate of growth were observed. A preliminary study of molds isolated from the tomatoes showed that some molds produced an antibiotic which inhibited some strains of *B. coagulans*.

These findings suggest that holding tomatoes after harvesting, particularly at relatively high temperatures, may be a predominant factor in the development of "flat-sour" spoilage.

MICROBIAL SPOILAGE of tomato products is generally associated with specific types of microorganisms, such as lactobacilli, yeasts, and certain butyric anaerobes, that tolerate a relatively high concentration of acid. Another type of spoilage, especially in canned tomato juice and whole tomatoes, known as "flat-sour," results from the development of *Bacillus coagulans*. Although a considerable amount of research has been conducted on this type of spoilage, sporadic outbreaks continue to occur. These outbreaks are unpredictable and losses to the canner are often severe. Since the organism involved produces little or no gas, spoilage cannot be detected until the cans of juice are opened. Thus, it is virtually impossible to segregate unopened cans of flat-sour juice from normal juice.

The term "flat-sour" is used rather loosely by many to describe any juice with an off flavor. Over a period of years this laboratory has examined many samples of abnormal tomato products. These products frequently had "off flavors" and the pH was below the normal range. Studies indicated that in many instances spoilage had developed before processing by such microorganisms as lactobacilli and yeasts. These organisms were destroyed by the heat process, but the by-products of growth were carried through to the processed tomato juice. Still another type of spoilage has been attributed to *Leuconostoc mesenteroides*. The presence of this organism in tomato juice usually indicates contamination of the product following the heat process, likely from contaminated cooling water and/or faulty can seams. This latter type of spoilage resembles "flat-sour" tomato juice in one respect in that the organism produces little or no gas and the ends of the cans remain flat. The pH of such juice is usually more acid than normal.

The true "flat-sour" juice is characterized by a disagreeable pungent odor and a bitter to sour flavor. Frequently, the pH of "flat-sour" juice is within the range for normal tomato juice. The organism responsible was described by Berry (1933) as a heat-resistant, acid-tolerant, spore-forming mesophile. He named the organism *Bacillus thermoacidurans*. Later studies by Smith *et al.* (1946) showed, however, that the cultural characteristics were the same as described for *Bacillus coagulans*, named by Hammer (1915).

Bacillus coagulans is extremely difficult to grow under acid conditions. Stern *et al.* (1942) found that repeated transfers in tomato juice were necessary to acclimatize the organism to the substrate so that spoilage would always develop when the organism was inoculated into normal juice.

Pederson and Becker (1949), using tomato juice of various pH values, showed that vegetative cells of more than one-third of 22 strains of *B. coagulans* grew and produced a typical flat sour in juice with a pH value of 4.21 or lower. Spores of the same 22 strains did not develop in juice of initial pH value lower than 4.32.

Rice and Pederson (1954), in a study of the effect of spore concentration on the growth of *B. coagulans*, found that as the spore concentration was reduced the minimum pH of tomato juice at which growth could occur rose. They also found that the amount of acid that could be tolerated by *B. coagulans* varied with the strain used. At a low spore concentration (20 or less per ml inoculum), only 2 of the 20 cultures

[a] Contribution No. 97, Plant Research Institute, Department of Agriculture, Ottawa, Canada.

[b] Present address: Emergency Supply Planning Branch, Department of Defence Production, Ottawa.

used were able to grow at pH 4.31 or lower. These same authors, studying the inhibitory influence of acidic constituents of tomato juice on the growth of *B. coagulans*, found that pH exerted the greatest effect.

Desrosier and Heiligman (1956) found that spores of *B. coagulans* were capable of germinating and growing in tomato juice if they were inoculated immediately after heat treatment or if the pH of the juice was increased. They also found that spores of *B. coagulans* caused spoilage in a juice buffered with potassium phosphate and adjusted to its original pH.

In a recent paper Krock *et al.* (1959) noted that the inhibition of *B. coagulans* by tomato juice was apparently a pH effect. They found the lowest pH at which spores of *B. coagulans* germinated was 4.30 at 37° C. At pH 4.35 spores germinated at 20 and 37° C, but not at 55° C. A pH of 4.82 was required for spore germination at 55° C.

The present study was undertaken to determine the incidence of flat-sour bacteria in canning areas across Canada; the effect ot pH on the development of *B. coagulans* on fresh and stored tomatoes and in processed tomato juice; and the nutritional requirements of *B. coagulans* isolated from material from the above survey. Results of these nutritional studies have been published elsewhere (Humphreys and Costilow, 1957).

EXPERIMENTAL

Incidence of flat-sour bacteria in canning areas. Since the natural habitat of *B. coagulans* is the soil, it follows that the organism would be carried to the processing plant on soil adhering to the tomatoes. All tomatoes arriving at the processing plant are subjected to washing either in a soak tank or by spray or by a combination of both methods. Thus, to determine the incidence of flat-sour bacteria from tomatoes from different areas, water from tanks used to wash the tomatoes was analyzed. The difficulty of shipping samples of water was overcome by a method developed at this laboratory—squares of heavy blotting paper (Kodak, extra, No. 120) that take up about 3 g of water. Preliminary tests showed that vegetative cells disappeared rapidly on drying but that spores survived for long periods.

The blotting papers were distributed to Government inspectors with instructions to dip each paper for 30 seconds in the wash water, air dry, and place in an envelope provided for shipment to Ottawa. Each plant received 9 papers, 3 to be dipped at the beginning of the canning season, 3 about the middle, and the remaining 3 toward the end. Instructions also stated that the dipping should be done at a reasonable time after the plant had started operations for the day.

The number of flat-sour bacteria on each paper was determined by placing the square of paper in a sterile jar containing 210 ml sterile water. This was shaken vigorously for 20 minutes in a Precision Scientific horizontal mechanical shaker, breaking up the paper to provide a homogeneous suspension. The initial dilution ratio was 1:70. Thus, portions of this suspension of 5, 2, and 1 ml respectively represented $\frac{1}{14}$, $\frac{1}{35}$ and $\frac{1}{70}$ ml of the original wash water. Suspensions were plated on thermoacidurans agar and incubated 48 hours at 45° C. Colonies developing on this medium were confirmed as flat-sour by transferring either all or a representative portion of the colonies to tubes of thermoacidurans broth containing an indicator and fermentation vials. Development of acid without gas was interpreted as positive indentification for flat-sour bacteria.

Determination of pH. Since acidity plays a predominant role in the development of *B. coagulans*, studies were started in 1953 and continued each year to determine the effect of maturity and of holding after harvesting on the pH of tomatoes. The method was varied from year to year, but essentially consisted of determining the pH of individual macerated tomatoes. In some instances the tomatoes at various pH levels were processed into juice and inoculated with cultures of *B. coagulans*, and pH was determined at various intervals of incubation.

Tomatoes for pH determination were selected at random and, visually, were completely red. Stored tomatoes were held at room temperature for 1–8 days. All determinations were made on a Beckman Model H_2 pH meter.

Pure culture studies. The cultures of *B. coagulans* used for inoculating the fresh tomatoes and tomato juice, with their sources, were: 43-P, National Canners Association; 56-186 A, National Canners Association; M-23, American Type Culture Collection (N.C.A. strain); 7050, American Type Culture Collection; 1604, isolated at this laboratory from a proven outbreak of flat-sour spoilage. Following preliminary trials on a number of media, test cultures were carried on a carbohydrate-free Bacto-Penassay Base agar (B270).

Inoculation of fresh and stored tomatoes. During one season, several bushels of fresh mature tomatoes were harvested on one day and held at room temperature. At intervals of 1, 2, 3, 6, and 7 days after harvest, one bushel of these tomatoes were spread out and inoculated with a suspension of the various cultures of *B. coagulans*. One bushel was left uninoculated as a control. Inoculation was done with a power spray, the spraying continued to "run-off." Each day, platings were made of tomatoes, selected primarily for size, from each bushel, including the control. The analysis consisted of weighing 100 g of tomatoes into sterile jars and adding 100 ml sterile water. This first dilution was shaken 5 minutes in a mechanical shaker, and further serial dilutions were prepared with 90-ml blanks of sterile water. From each dilution, 1.0-ml quantities were plated on thermoacidurans agar. Plates were incubated 48 hours at 45° C. Typical colonies of *B. coagulans* were counted, and the rate of growth noted.

RESULTS AND DISCUSSION

Incidence of flat-sour bacteria in tomato canning areas. Squares of paper dipped in water in tomato wash tanks were returned for analysis by about half of the plants for the 3 periods requested; the remaining plants returned one or two sets. Analysis of these papers (Table 1) showed that *B. coagulans* was present in all canning areas across Canada, with no evidence of unusual concentration in any particular area.

Of 268 papers analyzed, 214 (80%) showed counts of 100 or fewer spores of *B. coagulans* per ml of wash water. Only 12 papers (4.5%) showed spore counts over 500 per ml wash water. These results suggest that the concentration of spores of *B. coagulans* in processing-plant wash water was not heavy during the season when this survey was made. Also, during this particular season there were no reported outbreaks of flat-sour spoilage.

Relation of pH and susceptibility of tomato juice to spoilage. It is an accepted fact that pH is a predominant factor in the development of *B. coagulans* in tomato juice. The normal pH range of tomato juice, according to the Canned Food Reference Manual (1949), is 3.8–4.5, averaging 4.2. Early studies at this laboratory showed that tomato juice with pH values as high as 4.35 would not support the growth of many strains of *B. coagulans*. Studies with culture No. 1604, *B. coagulans* isolated from an outbreak of flat-sour spoilage, using a tomato juice with a pH of 4.20, showed a marked decrease in numbers in the early storage period. For vegetative cells this de-

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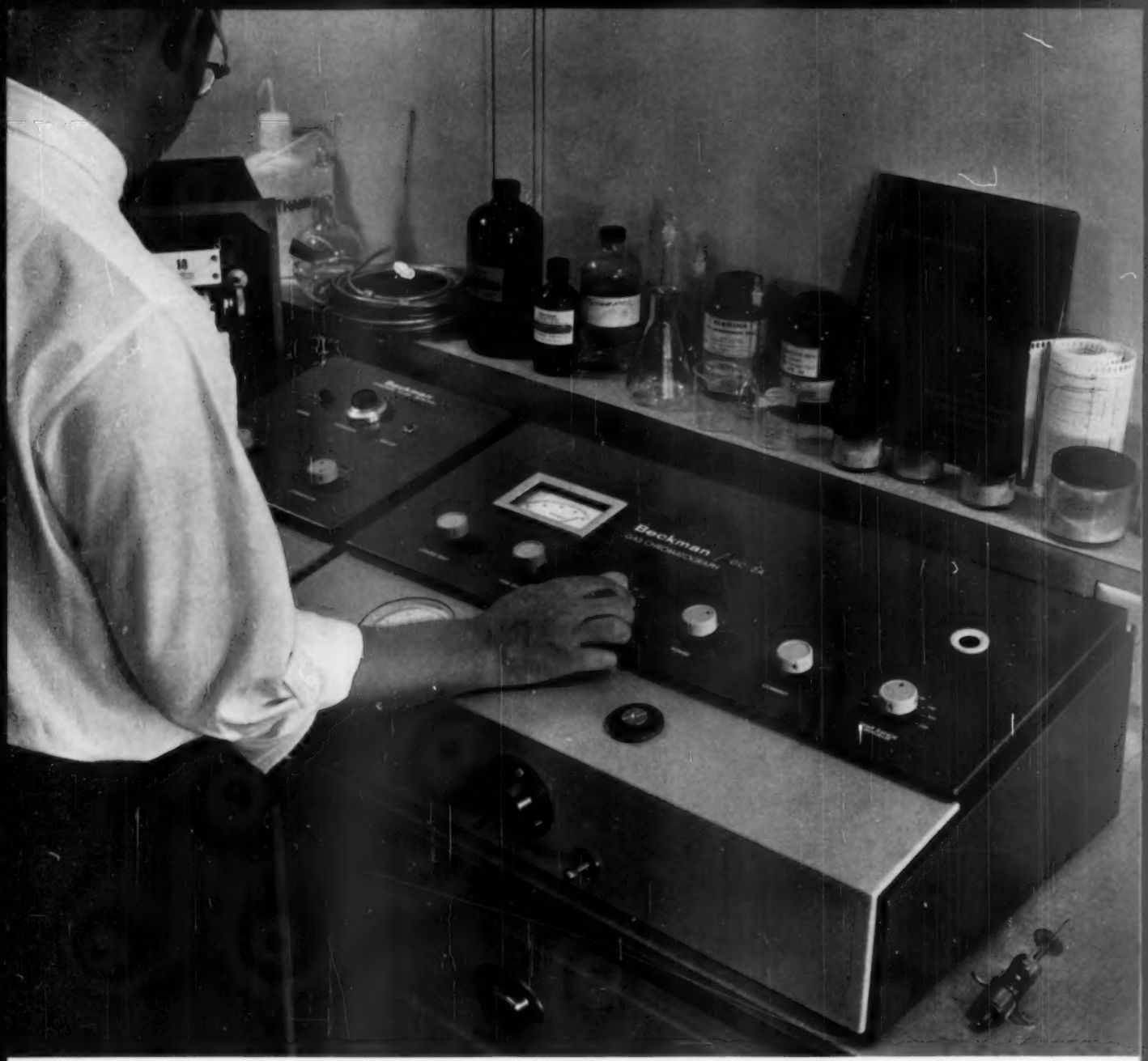

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Table 1. Distribution of Bacillus coagulans in canning plant wash waters in relation to area and period of season

Range of counts (/ml wash water)	Western Ontario		Eastern Ontario		All Ontario		Quebec		British Columbia	
	No.	%	No.	%	No.	%	No.	%	No.	%
Early season (Aug. 15–Sept. 15)										
0–100	31	81.6	34	65.4	65	78.2	17	85.0	8	72.7
101–500	7	18.4	10	19.2	17	18.9	3	15.0	2	18.2
501–1000			6	11.5	6	6.7			1	9.1
Over 1000			2	3.8	2	2.2				
Late season (Sept. 16–Oct. 15)										
0–100	53	86.9	39	86.7	92	86.8	18	66.7	14	100
101–500	8	13.1	4	8.9	12	11.3	8	29.6		
501–1000			2	4.4	2	1.9	1	3.7		
Over 1000										
Summary (over-all)										
0–100	84	84.8	73	75.3	157	80.1	35	74.5	22	88.0
101–500	15	15.2	14	14.4	29	14.8	11	23.4	2	8.0
501–1000			8	8.2	8	4.1	1	2.1	1	4.0
Over 1000			2	2.1	2	1.0				

crease was followed by a leveling off, and the count remained relatively constant during the remainder of the 10-day period. Spores continued to decrease throughout the entire storage period.

Studies both at this laboratory and by other workers (Becker and Pederson, 1950; Pederson and Rice, 1954; Stern *et al.* 1942) indicate that normal tomato juice does not provide a favorable medium for the growth of *B. coagulans*. The results of one series of studies to prove this conclusion are presented in Figs. 1 and 2. These data indicate that media using filtrates

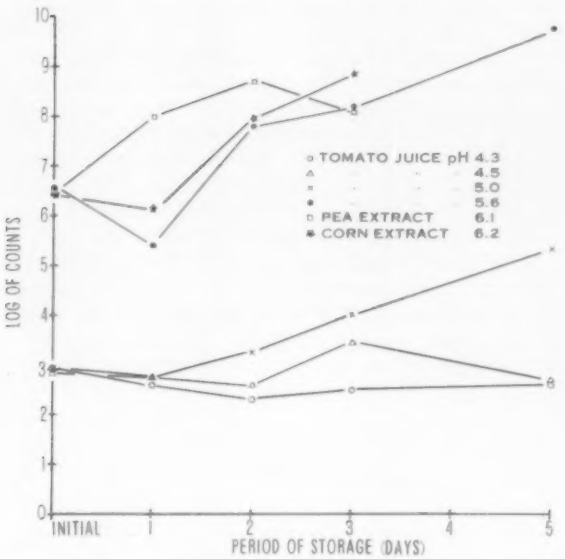

Fig. 1. Rate of growth of vegetative cells of Bacillus coagulans in tomato juice and in vegetable extracts.

from either corn or peas or from tomato juice adjusted to pH 5.5 provided more favorable substrate to support the growth of the test organism (43-P: National Canners Association Strain) than tomato juice unadjusted (pH 4.3) or juice adjusted to pH 4.5–5.0. At pH 5.0, however, there was a steady increase in numbers over the 5-day period. It is of particular interest to note the initial counts of vegetative cells in the various substrates (Fig. 1). All of these substrates

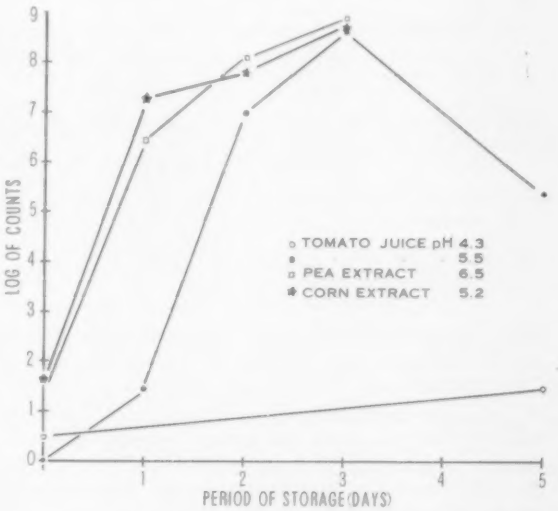

Fig. 2. Rate of growth of spores of Bacillus coagulans in tomato juice and in vegetable extract.

were given the same amount of inoculum from the same suspension. The data indicate that there was an immediate destruction of *B. coagulans* in the media with the lower pH values.

The initial marked decrease in numbers of vegetative cells of *B. coagulans* may be associated with the inhibitory properties of tomato juice as indicated in studies of Conner (1946) and Stern *et al.* (1942). The initial spore count also showed a decrease in the tomato substrates (Fig. 2), but this decrease was not as pronounced as that noted with the vegetative cells. The series of studies on spores (Fig. 2) suggests that the pH of the substrate was not responsible for inhibition, since the corn-extract medium had a lower pH than the tomato juice adjusted to pH 5.5. The data show a rapid growth of the test organism in the corn extract but definite inhibition in the tomato juice adjusted to pH 5.5. The inhibition did not last, however, since after 3 days the numbers of organisms in both corn extract and the pH 5.5 tomato juice were comparable.

pH values of fresh and stored tomatoes. The fact that pH has been found at this and other laboratories to be a predominant factor in the growth of *B. coagu-*

lans and in the development of flat-sour tomato juice suggested that a survey should be made of the pH of freshly harvested tomatoes and of tomatoes held in storage for a period comparable to that which tomatoes might be held at the processing plant.

Data on the pH of tomatoes over 5 years showed wide variations in the pH of fresh tomatoes from day to day during the period of harvest. There was also an indication that tomatoes became less acid as the season progressed. In 1957, for example, the average pH of apparently mature tomatoes harvested early in the season was 4.16; 7 days later the average pH was 4.40. Similar differences were noted in the range of pH values; the early tomatoes showed a range of 4.10–4.25, whereas tomatoes 7 days later showed a range of 4.27–4.60.

Considerable variation was observed from year to year, as indicated by the data in Table 2. These data

Table 2. Summary on 5-year study of pH of fresh tomatoes

Year	Average pH	Range of pH	No. tomatoes examined
1954	4.23	3.98–4.45	200
1955	4.34	4.11–4.60	100
1956	4.35	4.10–4.71	150
1957	4.32	4.10–4.75	175
1958	4.25	4.19–4.32	100

show that average pH during the peak of the harvest season was as low as 4.23 in one year and as high as 4.35 another year. This is also reflected in the range of pH; one year showed a pH range of 3.98–4.45, whereas the year with the highest average showed a range of 4.10–4.70. This indicates that in some years the pH of tomatoes may be within the range favoring the development of flat-sour bacteria.

Studies at one commercial plant on the effect of holding tomatoes before processing gave results of considerable significance. Holding harvested tomatoes either in the field or at the processing plant is common practice to develop a better color and/or is necessitated by the volume of tomatoes arriving at the plant. From the data in Table 3 it will be observed

Table 3. pH of fresh tomatoes held at room temperature at a commercial processing plant

Storage (days)	Av. pH	Range of pH
Inital	4.25	4.19–4.32
1	4.27	4.25–4.31
2	4.32	4.27–4.35
5	4.35	4.30–4.40
6	4.41	4.30–4.42
7	4.60	4.45–4.95
8	4.70	4.58–5.00
9	4.74	4.50–5.00
10	4.70	4.50–4.80
11	4.77	4.60–4.95

that the average pH of tomatoes increased from 4.25 to 4.77 during 11 days; the range was 4.19–4.32 to 4.60–4.95. It will also be observed that average pH increased from 4.41 to 4.77 from the 6th to 11th day. These pH values would permit the rapid growth of *B. coagulans*, with resultant flat-sour spoilage. During this period the pH of some tomatoes reached a high of 5.00.

Growth of B. coagulans on fresh tomatoes held at room temperature. Studies were made to determine the rate of growth of *B. coagulans* on tomatoes stored at room temperature. As indicated in the section on experimental procedure, fresh and stored tomatoes were sprayed at selected intervals with a suspension of two cultures of the test organisms (1604 and 43-P).

The data in Fig. 3 show that, with one exception (tomatoes inoculated on the 7th day of storage), num-

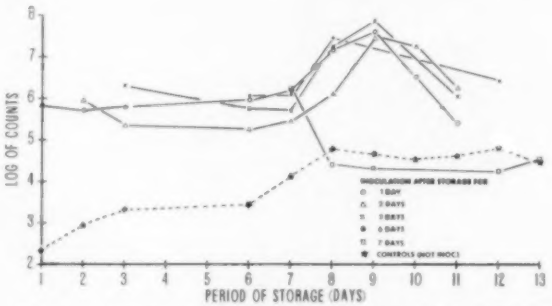

Fig. 3. Growth of Bacillus coagulans on fresh tomatoes stored at room temperature.

bers of the test organism increased markedly on the 8th day and continued to increase through the 9th day. This was followed by a steady decrease, which may be explained by the appearance of extensive mold contamination during this period. These molds competed with the test organism for food material, and subsequent tests showed the presence of growth-inhibiting substances. Extracts from 2 species of contaminating molds showed marked inhibitory properties against 2 of 5 cultures of *B. coagulans*.

Concurrently with the microbiological analyses, pH determinations generally showed a marked increase in pH value on the 8th day of storage (Fig. 4). Read-

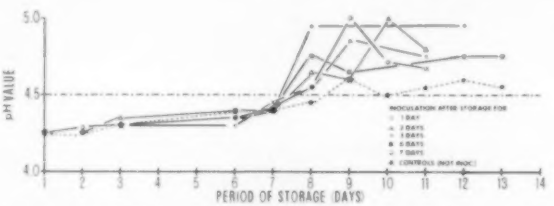

Fig. 4. Changes in pH values of fresh tomatoes spray-inoculated with Bacillus coagulans and stored at room temperature.

ings as high as pH 5.0 were noted on the 9th and 10th day with tomatoes inoculated on the 1st and 2nd days of storage, respectively. Tomatoes inoculated on the 3rd, 6th, and 7th days of storage reached respectively peak pH values of 4.85, 4.95 and 4.75. Control tomatoes reached a peak pH of 4.60 on the 9th day. No reason can be advanced for the lower pH value of the control tomatoes. All tomatoes were stored in the

same location, and thus were subjected to the same storage conditions.

The results of this study provide further evidence that the pH of tomatoes is an important factor on the rate of growth of *B. coagulans*. Regardless of the day of inoculation, the peak load of the test organism occurred during the period when the pH of the tomatoes was above 4.5.

Development of B. coagulans in tomato juice prepared from stored tomatoes. Tomato juice prepared from stored tomatoes of various pH values (4.25–4.95) was processed and inoculated with two cultures of *B. coagulans* (1604 and 43-P) and incubated 2 weeks at 45° C. The results (Table 4) show no change

Table 4. Development of Bacillus coagulans in tomato juice prepared from stored tomatoes at various pH levels

Period of storage	Inital pH of juice	Inoculated juice			
		Org. 1604		Org. 43-P	
		Final pH	Observations	Final pH	Observations
Fresh	4.25	4.25	No change	4.25	No change
2 days	4.30	4.40	Pellicle formation not flat-sour	4.35	Slight pellicle formation not flat-sour
5 days	4.35	4.40	Slight pellicle formation not flat-sour	4.40	No change
6 days	4.45	3.90	Heavy pellicle flat-sour	4.10	Heavy pellicle flat-sour
7 days	4.60	3.75	Heavy pellicle flat-sour	3.98	Heavy pellicle flat-sour
10 days	4.95	3.90	Heavy pellicle flat-sour	4.20	Heavy pellicle flat-sour

Incubated 2 weeks 45° C.

in juice at pH 4.25 prepared from fresh tomatoes; at pH 4.30 and 4.35 a surface-growth pellicle developed but the characteristic flat-sour odor was not detected. In tomato juice at pH 4.45, 4.60, and 4.95 the test organism showed rapid growth, with typical flat-sour spoilage.

Acknowledgments

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Studies on English (Persian) Walnuts, *Juglans regia*. III.

Stabilization of Kernels*

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Diamond Walnut Growers, Inc.[b]

(Manuscript received June 24, 1960)

SUMMARY

The relative stabilities of untreated and antioxidant-treated shelled walnuts were compared at moisture levels near the optimum range. Samples were maintained in air-packed, sealed glass jars exposed to ambient temperatures and incident lighting. Kernel stability was estimated by a panel of 10 experienced tasters as well as by measurements of peroxides, free fatty acids, and oxygen absorption. During the first 5 months no significant organoleptic differences were apparent between untreated and antioxidant-treated kernels containing 2.8–4.0% moisture. During the second 5 months untreated kernels held at 2.8% moisture became unacceptable and only small organoleptic changes were observed among the kernels containing 3.1–4.0% moisture. In general, all untreated kernels became unacceptable after 10 months. Among the untreated kernels, those having a moisture content of 3.1% exhibited the greatest stability. All antioxidant-treated kernels containing 2.8–4.0% moisture remained within acceptable limits for 18 months. Maximum stability was exhibited by kernels containing 3.3% moisture and treated with antioxidant II. The optimum moisture range observed in the present study for kernels held under ambient conditions was essentially the same as for those maintained in the dark at constant temperature (Swarthout *et al.*, 1958).

SHELLED WALNUTS are highly susceptible to darkening and the development of rancidity. Moisture, heat, light, and air are primary factors in their deterioration (Musco and Cruess, 1954, Rockland, 1955, 1957a, Rockland *et al.*, 1956, Swarthout *et al.*, 1958, Tappel *et al.*, 1957). It has been reported that walnuts are most stable at an optimum moisture content, above and below which they deteriorate at a more rapid rate (Rockland, 1955, 1957a, b, Rockland *et al.*, 1956). Under conditions of accelerated aging (140°F), maximum flavor and color stability were observed between the approximate limits of 1.4 to 4.5% moisture (Rockland, 1957a). At 95°F the optimum range was observed within the more precise limits of 2.0 to 4.0% moisture. At 38°F kernels were stable over a broader moisture range extending from about 1.5 to 6% moisture. It is now known that the optimum moisture varies nearly linearly with temperature between 38° and 140°F (Rockland, L. B., and A. A. Johnson, unpublished data). At 72.5°F it lies between 3.0 and 3.8% and coincides with minimum values of Rockland's $\Delta M/\Delta rh$ functions (Rockland, 1957b), which were derived independently from hygroscopic equilibrium data. Enhanced stabilization of kernels was observed when they were adjusted to within the optimum moisture range and treated with various antioxidants (Rockland, L. B., and J. C. Underwood, unpublished data). Swarthout *et al.* (1958) estimated the relative stabilities of antioxidant-treated kernels in the range of 3.0 to 5.0% moisture at moisture intervals of 0.5%. It was observed that oxygen absorption was minimized in both treated and untreated kernels when they were held at moisture levels of 3.0 to 3.5% in sealed cans.

The present studies were undertaken: to determine the stability of kernels in sealed glass jars under incident light and with moistures adjusted to circumscribe the optimum moisture range, and to estimate the relative effectiveness of two antioxidant formulations. Antioxidant I, a commercial product containing 20% butylated hydroxyanisole (BHA), 20% butylated hydroxytoluene (BHT), and 60% vegetable oil, had been evaluated in in-the-dark studies (Swarthout *et al.*, 1958). Antioxidant II contained multiple antioxidants and chelating agents in order to obtain maximum synergistic effects and to minimize the amounts of any one ingredient in the formula, as shown in Table 1. (Individual manufacturers should consult the U. S. Food and Drug Administration, Washington 25, D. C., and the food and drug officials of the individual states involved, to determine if the use of any proposed additive is permissible, and if so, what limitations are placed on its use.) The criteria employed to evaluate kernel quality included: organoleptic evaluations, peroxide values, oxygen absorption, and free fatty acids.

Table 1. Antioxidant II formulation.

Component	Per cent of	
	Solution	Total solids
Dipotassium salt of ethylenediamine tetraacetic acid	0.024	0.20
Citric acid	0.048	0.40
Butylated hydroxyanisole (BHA)	0.24	2.0
Butylated hydroxytoluene (BHT)	0.24	2.0
Propyl gallate	0.24	2.0
Sorbitan monostearate (Span 60)	0.52	4.4
Polyoxyethylene stearate (G-2147)	5.12	43.0
Polyoxyethylene sorbitan tristearate (Tween 65)	5.47	46.0
Isopropyl alcohol (91%)	88.1	....

* Presented at 20th Annual Meeting, Institute of Food Technologists, May 16, 1960, San Francisco, California.

[a] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

[b] Research Department, Diamond Walnut Growers, Inc., Stockton, California, with whom this work was conducted cooperatively.

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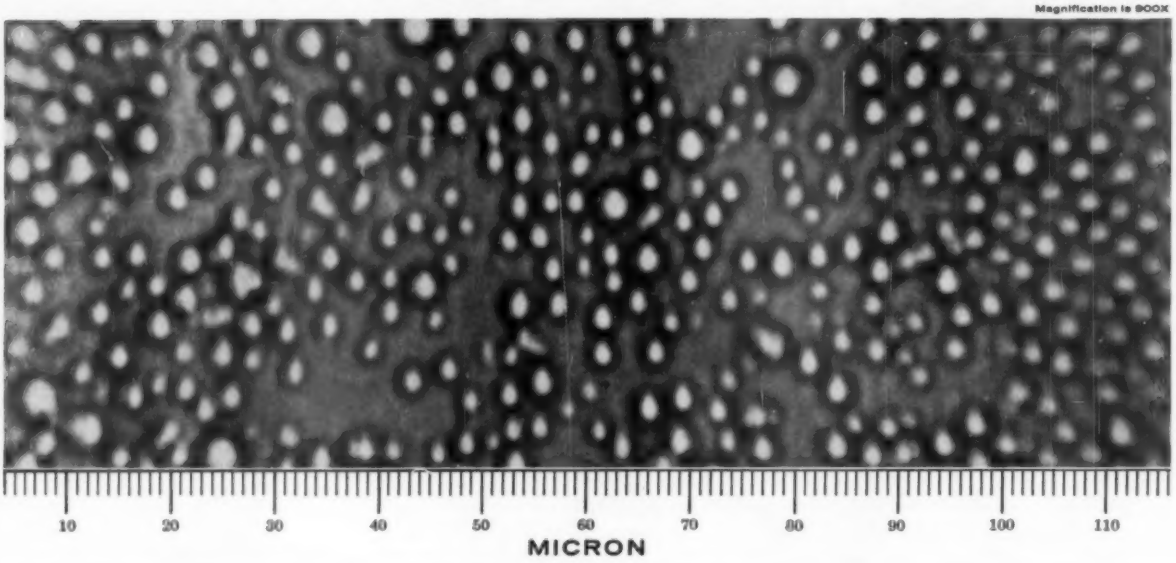

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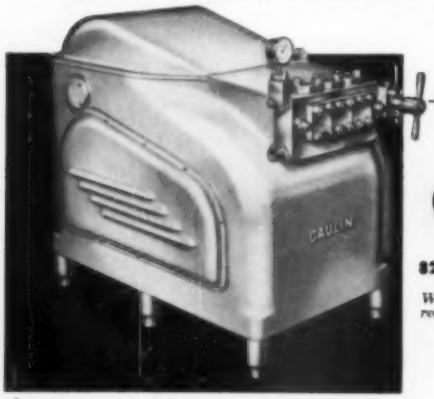

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EXPERIMENTAL METHODS

Walnut samples. Light-colored commercial-grade shelled walnut halves and pieces from the 1955 crop, were employed. The walnuts were machine-cracked and sorted for uniform size and color.

Antioxidant treatment. Approximately 105 lb of kernels were divided into three 35-lb lots, A, B, C. Lot A was untreated. Lot B was spread evenly in a single layer on a wire-mesh screen and sprayed uniformly on both sides with antioxidant I. The amount used was calculated to give a residual application (active ingredients) of 0.1% by weight, if 100% were utilized. Analysis of the applied antioxidant, using essentially the method of Anglin *et al.* (1956), indicated an efficiency of about 30% in the spray treatment, or an application of about 0.03% of total antioxidants. Lot C was treated by dipping kernels in a cheesecloth bag into antioxidant II. Kernels were allowed to drain on thick white blotting paper and exposed 24 hours to a current of moving air to remove isopropyl alcohol solvent. The treated kernels were then arranged in a single layer on clean white blotting paper and held an additional 24 hours at 100°F and 20% relative humidity to reduce the solvent residue further. The solvent odor appeared to be completely removed, but after the kernels were held for several weeks in sealed glass jars a slight alcoholic odor was readily detected in newly opened jars. However, after the nuts were removed from the jars, no detectable odor or flavor was detected by the taste panel. The kernels absorbed about 1% of their weight of solids, yielding an active antioxidant concentration of about 0.06%. Thus, the total antioxidant applied to Lot C was about twice that of Lot B.

Moisture adjustment. Moisture levels of the kernels were adjusted in a humidity- and temperature-controlled conditioning room. The kernels were held at a constant temperature of 71°F and the relative humidity was varied in accordance with the known moisture sorption isotherms (Rockland, 1957b) to obtain the desired moisture content. All lots were equilibrated 5 days at the lowest humidity. While they were maintained at the required equilibrium condition, 100-g samples from each of the three lots were weighed into 12-oz wide-mouth jars and sealed with rubber-gasketed metal lids. These samples were held in the conditioning room during subsequent equilibration of other samples at higher moisture levels. In sequence, the other samples were prepared similarly by equilibrating the kernels at successively higher pre-set humidities. Equilibration periods of 24–48 hours were allowed before the jars were weighed and sealed. Frequent moisture estimations during the hydration process aided in adjustment of moisture to the desired levels. The equilibrium relative humidities of the four lots of moisture-adjusted kernels averaged 26%, 30%, 45%, and 54% as measured with the electric hygrometer (Rockland, 1957b) at zero time and after 18 months.

Storage. Samples of all lots were held at 40°F until the initial quality evaluations were completed. These initial analyses were completed within 5 days of removal from the conditioning room. All samples were exposed to ambient inside daylight and temperature. In addition to exposure to daylight the samples were exposed to intermittent overhead fluorescent lighting. The ambient temperature varied over a range of 60 to 90°F during the 18-month period, averaging about 70°F. Ambient fluctuating light and temperature were employed rather than static conditions since it was known from previous studies that these conditions imposed a more severe stability test. Samples were removed for evaluation and analyses after periods of 0, 40, 135, 185, 280, 400, and 550 days. Moisture values did not change throughout the storage period.

ANALYTICAL METHODS

Moisture. Moisture was estimated by use of the Moisture Balance procedure described previously (Rockland, 1957b). (The mention of special instruments or materials does not imply endorsement by the U. S. Department of Agriculture over others of similar nature.) This procedure generally yields moisture values 0.2 to 0.5% higher than vacuum-oven procedures, but generally deviates less than 0.1% from Karl Fischer estimations. Xylene distillation and Moisture Balance procedures yielded almost identical values for the moisture content of six standard samples (Table 3). Low values obtained by the vacuum-oven procedure may be due to the hygroscopicity and rapid absorption of moisture by the vacuum-dried walnuts during operations required to obtain the final dry weight.

Table 2. Comparison of procedures for estimation of moisture.

Sample no.	% Moisture found			Moisture balance, % deviation from	
	Vacuum oven [a]	Xylene distillation [b]	Moisture balance [c]	Vacuum oven	Xylene distillation
1	2.87	3.13	3.13	0.26	0.00
2	3.05	3.13	3.25	0.20	0.12
3	3.74	4.17	4.25	0.51	0.08
4	3.92	4.27	4.28	0.36	0.01
5	3.69	4.07	4.17	0.48	0.10
6	3.88	4.27	4.33	0.45	0.06
			Av.	0.38	0.06

[a] Described by Rockland (1957b). Analyses performed on sliced kernels by M. McLaughlin, Fruit and Vegetable Chemistry Laboratory, Pasadena, California.
[b] Average of 3 replicate determinations.
[c] Procedure of Rockland (1957b).

Relative humidity estimation. A calibrated direct-reading A. C. Aminco-Dunmore electric hygrometer equipped with a multi-element humidity (10 to 99%) and temperature (40–100°F) sensing unit was employed as described previously (Rockland, 1957b).

Oxygen absorption. Oxygen absorption from the head-space gas was determined with the oxygen analyzer described by Swarthout *et al.* (1958). Since the samples were air-packed, the initial oxygen content of the head-space gas was 20.93%. Results are expressed as percentage of the original oxygen content absorbed, using the following relationship:

$$\text{Per cent oxygen absorbed} = \frac{20.93 - \%\ \text{residual oxygen}}{20.93} \times 100.$$

Free fatty acids. Walnut oil samples were prepared and analyzed for free fatty acids as described by Swarthout *et al.* (1958). Results are expressed as mg KOH/g oil.

Peroxide values. A 5-g sample of freshly expressed walnut oil was weighed directly into a 250-ml glass-stoppered Erlenmeyer flask. Fifty ml of a mixture of glacial acetic acid–chloroform (3:2) and 2.0 ml of a 50% solution of potassium iodide were added, and the flask was flushed with nitrogen, stoppered, and placed in the dark for 15 minutes. Fifty ml of water and 0.5 ml of starch indicator were added. The solution was shaken vigorously during titration with standardized 0.01*N* sodium thiosulphate.

Peroxide value (milliequivalent oxygen/kilo oil) =

$$\frac{\text{ml thiosulphate}^{e} \times N\ \text{thiosulphate} \times 1000}{\text{wt oil sample}}$$

Organoleptic evaluation. An experienced 10-member taste panel was employed to evaluate the flavor of kernels. Samples were rated on a hedonic scale of 10:

1. Excellent
2. Very good
3. Good
4. Fair
5. Slightly stale
6. Stale
7. Very stale
8. Slightly rancid
9. Rancid
10. Very rancid

During the analytical periods taste tests were conducted four times daily, two successive evaluations being conducted at about 10:30 a.m. and 2:30 p.m. Two test samples were presented for comparison with a pre-evaluated standard sample whose rating was made known to the panel. In addition, each participant was asked to rate the standard to determine if he

[e] Corrected for blank on reagents.

were in agreement with the assigned value. In this manner it was possible to maintain a running average of the standard sample, which became the assigned standard value for subsequent evaluations. Each sample was evaluated along with a different sample and compared with a different standard at each of the periods. One of the standard samples was chosen so that its flavor rating was equal to or higher than those of the test samples, whereas the second standard was chosen so that it was equal to or lower than the test samples. Large amounts of both standards were maintained at 0°F in small jars and removed as required. Each test sample was presented to each panel member twice and compared with each of two different test samples and standards. Statistical analyses of taste-test data yielded a standard deviation of 0.5 unit for the mean of twenty evaluations per sample. A rating of 5 (slightly stale) was arbitrarily set as the limit for acceptability.

Three or more replicate jars of kernels were taken for each series of analyses. Duplicate jars were used to determine the oxygen content of the headspace gas. Immediately after analysis of the headspace gas, the kernels were removed, ground, and crushed in a hydraulic press to express the oil employed for estimations of free fatty acids and peroxides. Separate samples were taken for organoleptic evaluations.

RESULTS AND OBSERVATIONS

Tables 3 and 4 respectively show the free fatty acid numbers and peroxide values observed for the cold-pressed oils obtained from control and antioxidant-treated kernels during exposure to ambient

Table 3. Free fatty acids in oils obtained from shelled walnuts; effects of moisture and time on acid number (mg KOH/g oil).

Time (days)	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II
	Moisture (%)			Moisture (%)			Moisture (%)			Moisture (%)		
	2.78	2.87	2.78	3.12	3.14	3.01	3.48	3.55	3.32	4.04	4.01	4.00
0	0.19	0.20	0.19	....	0.23	0.31	0.22	0.18	0.27	0.27	0.17	0.24
40	0.27	0.22	0.26	0.28	0.29	0.29	0.40	0.39	0.46	0.52	0.54	0.63
135	0.29	0.28	0.34	0.40	0.32	0.38	0.48	0.53	0.57	0.58	0.64	0.77
185	0.35	0.35	0.38	0.44	0.41	0.41	0.63	0.61	0.62	0.83	0.83	0.90
280	0.45	0.44	0.41	0.53	0.48	0.50	0.70	0.70	0.64	0.95	0.86	0.97
400	0.50	0.48	0.57	0.60	0.55	0.64	0.92	0.85	1.06	1.08	1.21	1.32
550	0.74	0.73	0.72	0.99	0.84	0.82	1.30	1.21	2.43	....	1.65	1.52

Table 4. Peroxide values of oils obtained from shelled walnuts; effect of moisture and time.

Time (days)	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II
	Moisture (%)			Moisture (%)			Moisture (%)			Moisture (%)		
	2.78	2.87	2.78	3.12	3.14	3.01	3.48	3.55	3.32	4.04	4.01	4.00
0	0.5	0.4	0.6	....	0.3	0.5	0.5	0.6	0.6	0.6	0.6	0.4
40	2.1	1.0	1.4	1.2	0.9	0.5	1.2	0.4	0.3	1.0	0.1	0.3
135	3.0	1.7	1.8	2.1	1.4	1.4	2.4	0.8	0.7	1.6	0.3	0.4
185	3.4	2.0	1.3	3.5	2.3	1.4	4.3	1.7	0.8	2.7	0.8	0.8
280	2.5	1.2	0.8	2.2	1.4	1.1	2.3	1.6	1.0	2.9	1.1	0.8
400	4.2	1.8	2.2	3.5	2.3	2.2	4.4	1.3	1.5	5.0	1.4	1.3
550	3.9	1.9	1.1	3.8	1.2	1.9	2.6	0.7	1.1	....	0.6	0.5

Table 5. Per cent of headspace oxygen absorbed by shelled walnuts; effect of moisture and time.

Time (days)	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II
	Moisture (%)			Moisture (%)			Moisture (%)			Moisture (%)		
	2.78	2.87	2.78	3.12	3.14	3.01	3.48	3.55	3.32	4.04	4.01	4.00
0	1	(−1)[a]	1	....	1	1	1	1	(−2)	0	0	(−1)
40	12	9	7	9	8	7	8	7	6	9	6	6
135	12	14	10	12	12	9	13	13	9	12	13	12
185	21	16	13	17	18	15	25	16	14	20	17	14
280	25	20	16	22	20	14	26	18	17	39	22	19
400	42	26	22	22	23	18	41	25	19	47	27	20
550	56	29	31	51	30	23	62	35	26	....	42	31

[a] The values enclosed by parentheses represent apparent enrichment of oxygen in the headspace.

Table 6. Organoleptic evaluations of shelled walnuts[a]; effects of moisture and time.

Time (days)	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II	No treatment	Antiox. I	Antiox. II
	Moisture (%)			Moisture (%)			Moisture (%)			Moisture (%)		
	2.78	2.87	2.78	3.12	3.14	3.01	3.48	3.55	3.32	4.04	4.01	4.00
0	4.0	3.9	4.0	3.4	3.6	3.6	3.6	3.8	3.8	3.8	3.7	3.7
40	4.0	4.7	4.7	4.0	4.5	3.9	4.2	4.6	4.2	4.4	4.4	4.4
135	3.6	3.9	3.5	3.5	3.4	3.7	3.2	3.6	3.2	3.7	3.8	3.6
185	5.1	4.5	4.2	4.2	4.2	4.4	4.4	3.9	3.8	4.5	4.0	4.1
280	6.0	5.1	5.2	5.6	5.1	4.8	5.4	4.7	4.5	5.3	4.4	4.4
400	5.4	4.8	4.4	4.7	4.4	4.3	5.6	4.9	4.4	6.1	4.3	4.4
550	7.7	4.3	4.8	6.0	4.7	4.7	7.3	4.2	3.7	8+	4.5	4.8

[a] Based on a hedonic scale of 10. The limit of acceptability was set arbitrarily at 5 (slightly stale).

light and temperature. The percentages of the original oxygen content of the headspace gas that had been absorbed in the various experimental lots are presented in Table 5. The values in Tables 3 to 5 represent the averages of two or more replicates. Organoleptic evaluations obtained before and during the extended holding periods are presented in Table 6.

Peroxides. The peroxide values observed for the two series of antioxidant-treated kernels were on the same order of magnitude and significantly lower than those of the untreated kernels throughout the 550-day period at the four moisture levels. In general, kernels were organoleptically acceptable when peroxide values remained below 2.5. Musco and Cruess (1954) reported that nut kernels were judged to be rancid when peroxide values reached 2.0. In the present study, however, acceptable kernels were observed with peroxide values greater than 4, whereas unacceptable kernels have been observed with peroxide values below 1.2. The imperfect correlation between peroxide values and organoleptic quality would appear to be due to the anomalous variations of peroxide values as a function of storage time.

Oxygen absorption. The initial rates of oxygen absorption by the untreated and antioxidant-treated kernels were essentially the same at all moisture levels. After 5–6 months, however, the untreated kernels held at 2.8, 3.5, and 4.0% moisture absorbed oxygen from the headspace gas at a higher rate than antioxidant-treated kernels at the same moisture levels. At 3.1% moisture the untreated kernels absorbed oxygen at about the same rate as the treated kernels for about 13 months, which suggests a critical moisture level of about 3.1% moisture for untreated kernels. Within the narrow range of 2.8–4.0% moisture, the moisture optimum appeared to be less critical for treated kernels, although in general the data suggest that oxygen absorption was slightly lower at 3.0 and 3.5% moisture.

The slight initial apparent enrichment in the oxygen content of the headspace gas of several antioxidant-treated samples is probably not significant. However, apparent oxygen enrichment in newly sealed tin cans and other tightly closed containers has been observed frequently in previous walnut stability studies.

Free fatty acids. The total free fatty acids increased generally with moisture content and time for the kernels from all lots. There was no clear direct relationship bteween the formation of free fatty acids and the development of rancidity, although there was a general tendency for the free fatty acids to increase as the kernel flavor decreased. The free fatty acid values observed for the untreated kernels were about 100% greater than those observed for kernels stored in the dark by Swarthout *et al.* (1958) for the same period at the same moisture content. However, the walnuts employed in the present study were obtained from a commercial lot of mixed varieties, whereas those used in the "dark" study by Swarthout *et al.* were a freshly shelled lot of the Payne variety.

Flavor. All of the samples remained acceptable for 135 days in sealed glass jars under ambient conditions. The control samples, which contained 3.0–4.0% moisture, remained acceptable for at least 185 days, whereas the sample at 2.8% moisture became unacceptable sooner. All of the antioxidant-treated kernels were acceptable after 18 months, although several lots experienced transient periods of apparent unacceptability (Table 6). Fig. 1 compares the flavor scores for untreated and antioxidant-treated kernels held at 3.4% moisture during the 550-day period. At 3.12% moisture the flavor of untreated kernels was optimum and oscillations in the flavor scores as a function of time were most apparent. Minimum changes in flavor were observed at moisture levels of 3.0–4.0% for kernels treated with antioxidants I and II, respectively. Flavor stability was slightly better in kernels treated with antioxidant II than in kernels treated with antioxidant I.

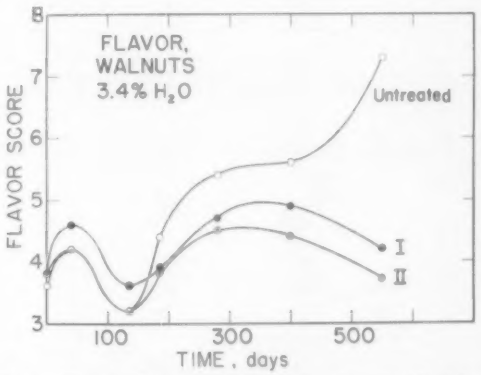

Fig. 1. Flavor changes in untreated and antioxidant-treated kernels containing 3.4% moisture. Open circles, Control. Solid circles, antioxidant I treatment. Double circles, antioxidant II treatment.

Inversions in flavor scores, as a function of time, have also been observed in previous unpublished studies. These inversions, however, were generally believed to be due to random errors or low precision in the organoleptic evaluations. In the present studies the flavor evaluations were repeated three times when apparently anomalous inversions were observed. These repeated evaluations yielded essentially identical results.

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Studies on English (Persian) Walnuts, *Juglans regia*. IV.

Belt-Trough Drying of In-Shell Walnuts

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AT AMBIENT temperatures, English walnut kernels are most stable when the moisture level is 3.5 ± 0.3% (Rockland, 1957 b; Rockland *et al.*, 1961 a). Moisture contents above or below the optimum level result in more rapid darkening of the kernel and accelerated development of rancidity. Drying in-shell walnuts to the optimum moisture range for kernels would be advantageous for the stabilization of whole nuts prior to shelling. In-shell drying would permit cracking and shelling to be conducted when the kernels have maximum stability and would eliminate the secondary abrasion from dehydration after shelling. The studies described here were undertaken to determine the utility of the belt-trough dryer for the dehydration of in-shell walnuts and to estimate conditions under which walnuts can be dehydrated most efficiently without impairing initial kernel quality.

A maximum drying temperature of 110°F was suggested by Batchelor and Christie (1924) for the dehydration of in-shell walnuts. Later work, however, indicated that they can be exposed to comparatively high temperature for short periods without harming initial quality (Rockland *et al.*, 1961 b). The work described herein includes final kernel temperatures in the range of 137–156°F, corresponding to drying air temperatures of 175–225°F.

RESULTS AND DISCUSSION

Variations in kernel moisture content and temperature as a function of drying temperature and time are shown in Table 1. No significant changes in kernel flavor, odor, color, or general appearance were observed during dehydration at 175 and 200°F or after equilibration in sealed containers for 8 days at ambient temperature. However, after the equilibration period, kernels dried for 15–20 minutes at 225°F had a good but slightly reduced flavor quality. It has been demonstrated (Rockland *et al.*, 1961 b) that quality is not impaired if kernel temperatures do not exceed 145°F during rapid drying. Since the kernel temperatures of walnuts dried at 225°F exceeded 150°F for 5–10 minutes it would be expected that storage quality may have been impaired, although it was not clearly apparent following the short holding period.

Kernel moisture levels were consistently lower for the samples equilibrated in-shell than for correspond-

Table 1. Dehydration of in-shell walnuts on the belt-trough dryer at 175, 200, and 275° F

Drying time (min)	Inlet air temperature (°F)	Exhaust air temperature, w b/d b (°F)	Kernel temperature (°F)	Moisture content (%, wet basis)		
				Kernel initial[a]	Kernel final[b]	Ground whole nut[c]
0	175	.....	80	4.39	4.39	.....
8		87/148	.....	.....	.....	.....
15		.....	.....	4.70	4.00	.....
30		85/155	133	4.38	3.90	6.15
45		.....	137	3.85	3.15	5.52
60		.....	134	3.57	3.10	5.22
0	200		80	4.39	4.39	.....
5			111			
8		90/163				
10			134	4.42	3.82	6.79
20			143	4.12	3.42	5.83
30		91/172	143	3.95	3.25	5.34
0	225		80	4.39	4.39	.....
5		95/184	115	4.87	4.25	7.67
10			149	4.57	3.85	6.81
15			153	4.52	3.68	6.00
20			156	4.60	3.32	5.73

[a] Kernels removed immediately after drying.
[b] Kernels removed after equilibration of whole walnuts for 8 days.
[c] Whole nuts equilibrated and ground for moisture estimations.

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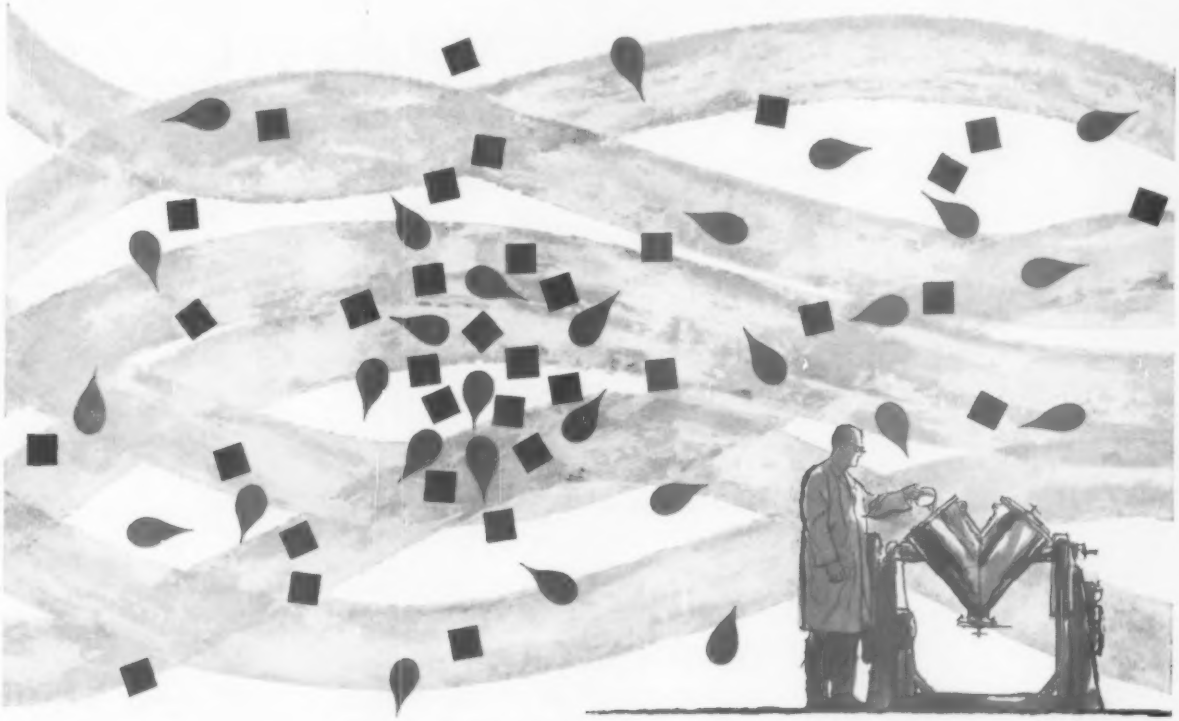

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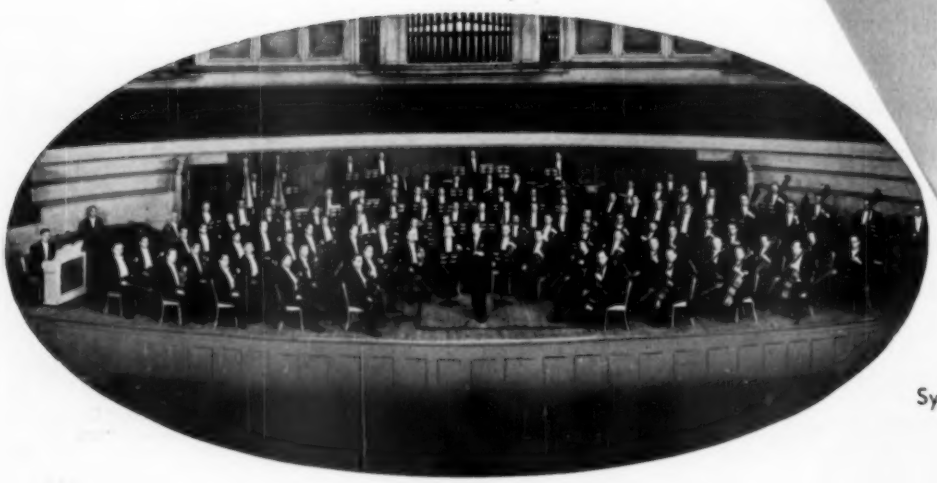

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ing samples shelled immediately on removal from the dryer. This indicates a transfer of moisture from the kernels to the shells during the equilibration period. At a given temperature, the sole requirement for such a transfer of moisture is that the shells be drier than the kernels, a condition certain to exist in the samples taken from the dryer. Shell materials account for about 50% of the total weight of a walnut but contain ⅔ of the moisture. The ratio of shell weight to kernel weight and the ratio of shell moisture to kernel moisture immediately upon leaving the dryer determine the equilibrium moisture of the unshelled samples after 8 days of storage.

It is apparent from the present studies that in-shell equilibration is essential for rapid dehydration of whole walnuts to the optimum kernel moisture without damaging kernel quality. The data indicate drying times well in excess of 30–40 minutes for the kernels shelled immediately upon removal from the dryer. From a quality standpoint, the half hour or longer drying time would be undesirable, particularly at the higher drying temperatures.

On the basis of the plotted data, optimum drying temperature was established at 200°F. At that temperature, kernel moisture dropped from an initial value of 4.4% down to 4.2% in 17 minutes. The samples taken from the dryer at the same time, and equilibrated in the shell for 8 days, showed a moisture level of 3.5%. Increasing the drying temperature to 225°F did not reduce the time required to dehydrate the kernels to the same equilibrated moisture level.

On the other hand, reducing the drying temperature to 175°F increased the drying time from 17 minutes to about 37 minutes for equilibrated walnuts.

No off-flavor was detected in any of the experimental samples. Color, odor, and appearance were also judged normal or good. Final kernel temperature was 137°F with 175°F air, 143°F with 200°F air, and 156°F with 225°F air.

The data for the samples shelled immediately upon removal from the dryer show a general increase in kernel moisture during the early stages of drying. This is a period marked by high shell temperatures and comparatively low kernel temperatures, giving rise to the possibility of moisture evaporating from the shell and condensing on the kernel. This would be a transient condition, existing only as long as the kernel temperature is at or below the dew-point temperature of the small pocket of air surrounding the kernel in the shell. The data indicate that this condition lasts about 15 minutes at 175°F and 5–6 minutes at 225°F. Sampling was inadequate for estimate of the transiency for drying at 200°F.

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A Study of Macaroni Products Containing Soy Flour*

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SUMMARY

Methods are presented for evaluation of macaroni products for per cent water absorption and per cent of solids and nitrogen extracted during cooking. These analytical methods supplemented by taste-panel evaluation and rat assay indicate that adding Ardex 550, a soy flour, to semolina at levels of 12.5, 17, and 25% increases the firmness of spaghetti subjected to long cooking periods and significantly improves the nutritional quality of macaroni products.

During the past several years there has been an increasing interest in protein and other nutritional supplements in basic economical food items. Simultaneously with these interests in nutrition, economics, and availability, came the problem of testing such products for uniformity and quality, especially for institutional feeding and the canning industry. Harris and Sibbitt (1958) reviewed the literature on methods of evaluating the cooking quality of macaroni products and presented data on the cooking properties of some new durum wheat varieties. Procedures for analyzing macaroni products were described in "Cereal Laboratory Methods" (Am. Assoc. Cereal Chemists, 1957). James J. Winston (1959) and Winston and Jacobs (1947) outlined a method for evaluating macaroni products that was approved by the U. S. Department of Agriculture and the National Macaroni Manufacturers Association. Winston (1959) reported results of cooking data for spaghetti and egg noodles produced by a number of different manufacturers. The data in the literature on the cooking quality of various macaroni products have been based on a specified cooking time such as 20 or 30 minutes under a set of conditions. This investigation was designed to study the functional and nutritional properties of macaroni products containing soyflour. The data on functional properties include patterns for water absorption and the quantities of solids and nitrogen extracted for a series of cooking times.

EXPERIMENTAL

Materials

The semolina spaghetti and elbow macaroni used in this study as control were commercial products purchased at different times and at several retail markets. The products containing different percentages of soyflour in combination with semolina were prepared by commercial manufacturers on an experimental basis. Used in all the experimental macaroni products was Ardex 550 soyflour (manufactured by Archer-Daniels-Midland Co., Minneapolis, Minnesota).

It was determined that commercial spaghetti made from 100% durum wheat was not uniform in diameter throughout the entire length. Representative materials for evaluation were obtained by selecting spaghetti of uniform diameter.

The diameter of the spaghetti used for analytical data was measured using a micrometer caliper with a maximum variation of ± 0.002 in. No attempt was made to measure the elbow macaroni, but uniform unbroken elbow macaroni was used for all the tests. The quality tests performed during this investigation were objective and subjective.

Analytical Methods

The methods for determination of water absorption and of solids and nitrogen extracted into the cooking water were:

Cooking. Several glass beads and the screen were placed in a 600-ml tall form beaker and tared within ± 0.1 g. 300 ml of hot distilled water were measured into the tall form beaker; a condenser (E. H. Sargent Co. condenser-reflux No. S-22742) was placed in position over the top of the beaker; the water was brought to a boil and refluxed until the top of the beaker was hot. A sample of macaroni product was weighed to 22.4 ± 0.1 g, using 3-in. lengths of the center 9-in. section of long spaghetti.

The sample was transferred quantitatively into the boiling water, with timing started immediately; stirred so all pieces were under water and not sticking together; and boiled at a moderate rate of reflux for varying periods.

Determination of Water Absorption. The beaker was removed from the hot plate, the timer was set for 5 minutes, the wire screen was inserted into the beaker so as to touch the screen on the bottom (Fig. 1), and the cooking water was drained rapidly into a tared 400-ml beaker. After most of the water had been drained, the beaker was agitated manually to remove the mechanically held water in the cooked sample. Draining was continued until no more water ran from the sample. The screen was removed while the beaker was tilted, so that any water on the screen did not run back into the beaker. The 600-ml beaker was turned upright, and the condensate was absorbed from the upper part of the beaker with a paper towel. The beaker was placed on a scale, and an approximate weight was taken. If water appeared in the bottom of the beaker, it was drained before a final weighing.

The cooked sample was weighed to ± 0.1 g at exactly 5 minutes from end of cooking (at 5 minutes, the sample had cooled and the error due to surface drying was minimized). The weight (a) minus tare (b) and weight of sample (22.4 g) equalled the grams of water absorbed:

$$\frac{(a) - (b) - 22.4}{22.4} \times 100 = \%\ H_2O \text{ absorption}$$

Determination of Extracted Solids. The cooking water collected in the 400-ml tared beaker was evaporated to dryness in a 130°C oven to constant weight, desiccated, and weighed to ± 0.01 g:

$$\frac{\text{Weight} - \text{tare}}{22.4} \times 100 = \%\ \text{solids extracted}$$

Determination of Extracted Nitrogen. The solids were transferred quantitatively from the 400-ml beaker with H_2SO_4 and water.

* Technical Paper No. 182, Archer-Daniels-Midland Company.

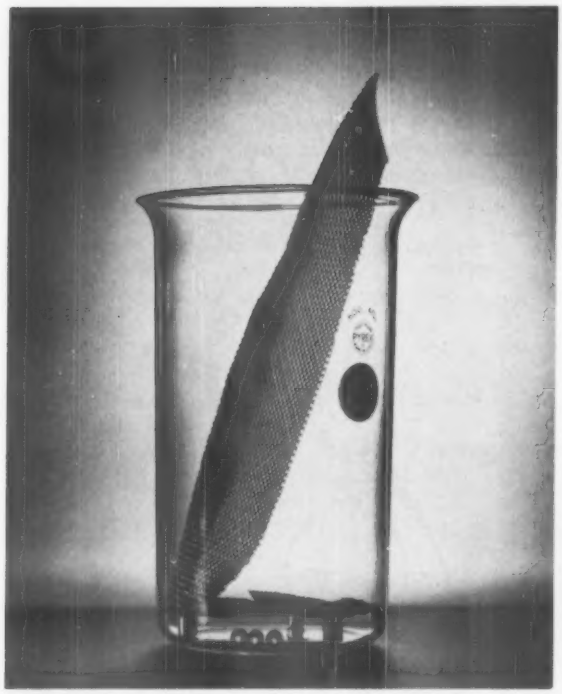

Fig. 1. Screens for 600-ml Berzelius tall form beaker.

Determine Kjeldahl nitrogen according to AOCS procedure (Am. Oil Chemists Soc., 1959).

Calculation:

$$\frac{\text{Total nitrogen}}{22.4} \times 100 = \%\ \text{nitrogen extracted}$$

Experimental Autoclaving Method

A 22.4-g sample of elbow macaroni was weighed into a 400-ml beaker; 300 ml of boiling distilled water were added during agitation. Samples were autoclaved immediately at 240°F for 30 minutes.

Experimental Canning Method

Test packs were produced at the Continental Can Company's Metal Division Research and Development Department Pilot Plant in Chicago (Continental Can Co., 1959). The method used for canning spaghetti in tomato-cheese sauce was as follows: The spaghetti was blanched for 10 minutes at 212°F. The blanched spaghetti was held in cold water until immediately before use; then it was drained. The 300 x 407 cans were filled with a measured amount of sauce as a cushion; 7 oz. of drained blanched spaghetti was added, and the fill completed with the heated sauce. The filled cans were closed at once and placed in the retort. The retort was exhausted by bringing the temperature up to 220°F over a 5-min period, then it was brought rapidly to 240°F and maintained there for 75 minutes. Two minutes were allowed for the temperature of the retort to drop to 212°F after the process was completed. Cans were water-cooled to 105°F. The pH of sauce and spaghetti in the finished product was 4.8 after the product had become equalized.

Three cans of each type of spaghetti were evaluated after storage at 70°F for 1, 2, 3, and 6 months, and at 100°F for 1 and 1.5 months.

Protein Assay Method

Used in this study was a 42-day rat-growth assay developed in the Archer-Daniels-Midland Company laboratories. Each lot contained 6 male weanling albino rats weighing 50–60 g at the start of the assay. The animals were individually caged in an air-conditioned room at 75°F. The animals had free access to water.

Initial and final weights were obtained from 3 consecutive daily weighings. Weights were also obtained at weekly intervals during the test. The rations were fed on a controlled-intake basis. The food was weighed fresh each day, and the rejected food from the previous day was discarded.

Food consumption, weight gain record, and protein (N × 6.25) efficiency were averaged weekly.

The experimental diet consisted of corn starch, non-nutritive cellulose, fat-soluble and B-vitamins, salt mixture-W (Osborn-Mendel-Wesson Mineral Mix, purchased from Nutritional Biochemicals Corporation, Cleveland, Ohio), raw clarified soybean oil, and sufficient protein assay material to supply a 10% level of protein in the diet. The diet was also adjusted to contain 8% of fat and 2% of fiber.

RESULTS AND DISCUSSION

In establishing procedures for determination of water absorption and solids and nitrogen extracted from macaroni products, it was demonstrated that the most important factors in obtaining repeatable results were sampling of the product to be tested and the method of de-watering after cooking.

Table 1 summarizes analyses for water absorption of a uniform sample of 17% Ardex 550-semolina spaghetti. The range in cooking times (10–90 minutes) was selected for this study as representative of the variation in heat processing encountered during canning operations and in institutional feeding.

Table 1. Summary of water absorption determinations for 17% Ardex 550-semolina spaghetti (0.070±0.002 in. in diameter).

Cooking time (min)	Water absorbed		
	g/22.4 g spaghetti	%	% difference (±)
10	27.2	121	0.42
10	27.4	122	
20	40.2	179	
20	40.6	181	
20	40.7	182	2.45
20	41.9	187	
20	42.1	188	
30	51.4	229	0.86
30	52.1	233	
40	57.3	256	0.39
40	57.8	258	
50	62.8	280	0.70
50	63.7	284	
60	68.5	306	0.16
60	68.8	307	
70	71.5	319	
70	72.3	323	1.7
70	73.9	330	
80	75.3	336	0.68
80	76.2	340	
90	76.8	343	
90	78.2	349	1.15
90	78.8	351	

Increased firmness of macaroni products containing soy flour has been demonstrated by autoclave experiments. Fig. 2 visually demonstrates the effectiveness

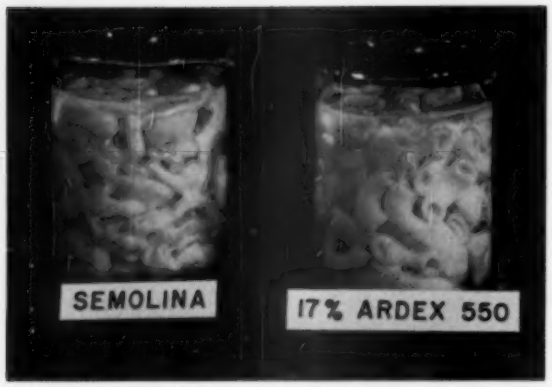

Fig. 2. A demonstration of improved firmness of autoclaved soy elbow macaroni.

of soy for improving resistance to overcooking when autoclaved 30 minutes at 240°F.

Fig. 3 shows typical water absorption patterns for semolina spaghetti of three different diameters, with cooking time as the variable. The per cent water absorption for spaghetti is inversely proportional to the diameter of the spaghetti and directly correlated with increased firmness in canning and institutional cookery.

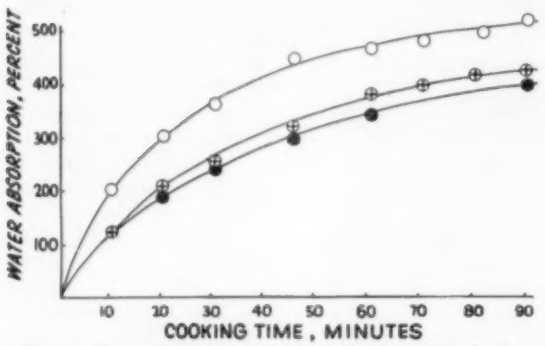

Fig. 3. Data showing water absorption patterns for semolina spaghetti of different diameters with cooking time as the variable. ○ 0.055 inch; ⊕ 0.065 inch; ● 0.075 inch.

Fig. 4 compares water absorption patterns of Ardex 550-semolina and semolina spaghetti for two common diameters. These data further show that soy-semolina blends for macaroni products decrease water absorption.

Fig. 5 shows patterns for per cent solids extracted in the cooking water. Note that the solids content in the cooking water is somewhat greater for a soy spaghetti than for 100% semolina spaghetti. These patterns are averages of various spaghetti irrespective of diameter.

The amount of solids and protein extracted during long cooking periods could be significant in the canning industry. Soy flour would contribute to a higher solids content in the cooking water since it contains about 35% soluble carbohydrate and 6–8% ash.

The pattern for solids extracted into the cooking water when cooking time was varied was about 2.5% greater for spaghetti containing soy flour than for a 100% semolina product. This is not considered significant from the standpoint of nutrition because the carbohydrates in soy are mostly pentoses, which are not utilized nutritionally.

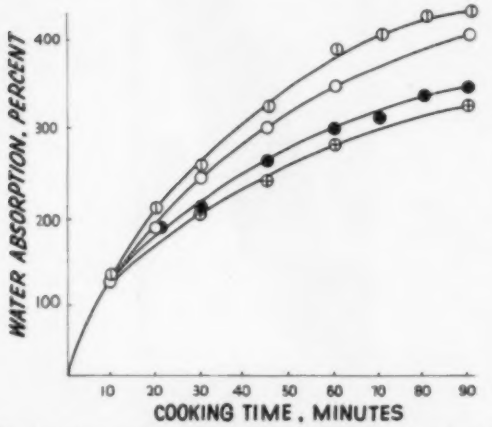

Fig. 4. Comparative water absorption patterns for Ardex 550 soy and semolina spaghetti with cooking time as a variable. ● 17% soy spaghetti, 0.065 inch; ⓘ semolina, 0.065 inch; ⊕ 17% soy spaghetti, 0.075 inch; and ○ semolina, 0.075 inch.

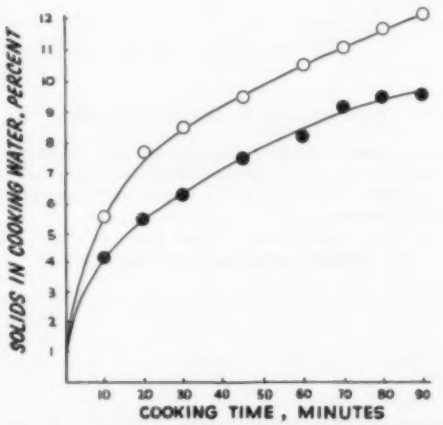

Fig. 5. Comparative patterns of solids extracted at various cooking times: ● semolina spaghetti; ○ 12.5 and 17% Ardex 550 soy spaghetti.

The native protein of soybean is completely water-dispersible. Therefore, the amount of protein extracted into the cooking water was determined for soy spaghetti and macaroni products. The data for protein extracted during various cooking times have been calculated as the per cent of the total protein (all data calculated as nitrogen). Table 2 summarizes the data on protein extracted from various types of spaghetti, with and without soy flour.

The percentage of the total protein in macaroni products that is extracted into the cooking water

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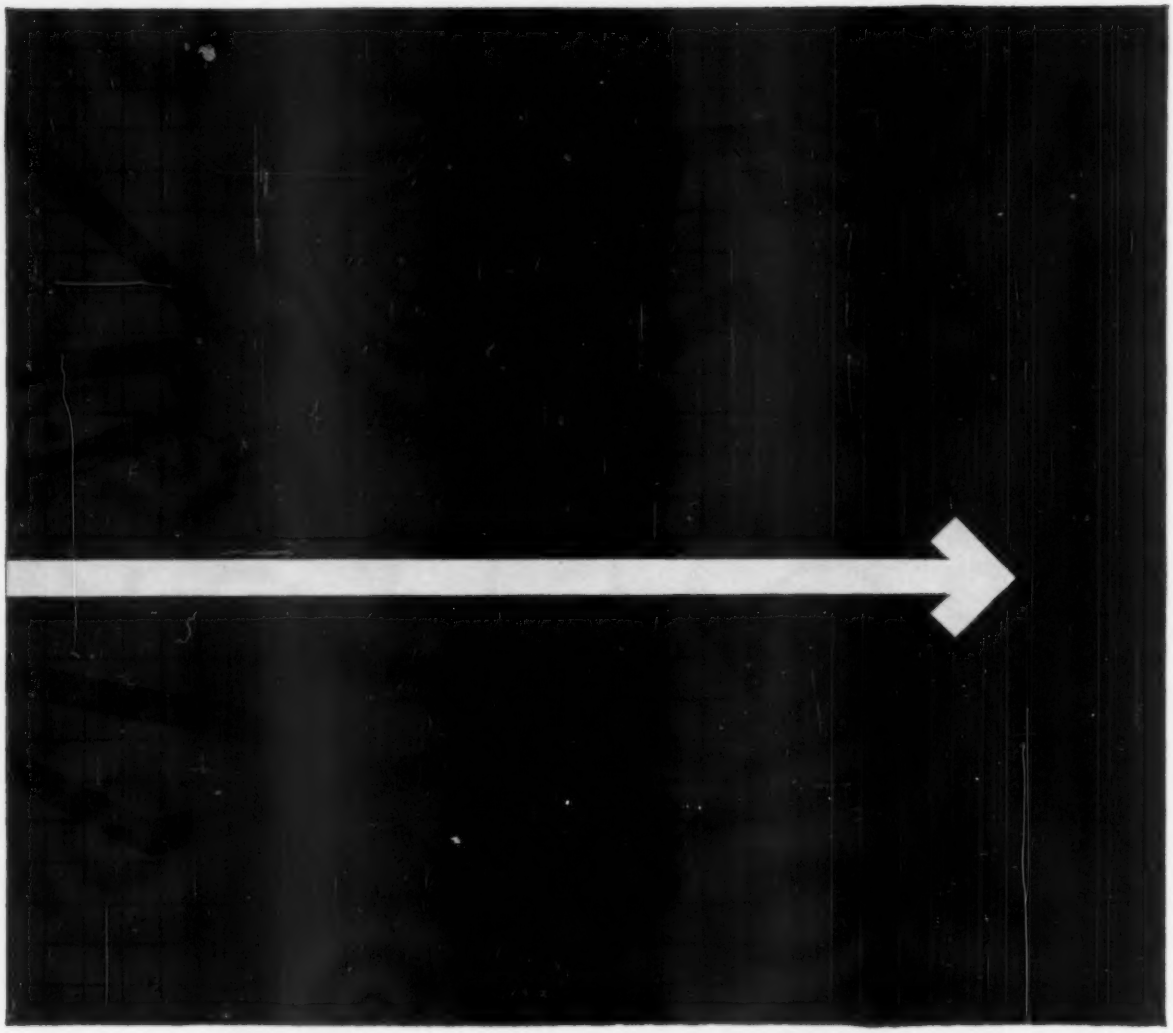

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Table 2. Nitrogen extracted into cooking water (% of total nitrogen).

Description	100% No. 1 Semolina		Ardex 550–Semolina[a]	
			17%	25%
Diameter (in.)	0.065	0.075	0.070	0.075
Total nitrogen	2.25	2.25	3.36	3.84
% of total nitrogen extracted in cooking time of:				
10 minutes	3.56	3.69	3.51	2.84
20 minutes	4.96	4.49	4.67	4.66
30 minutes	4.53	.....	.....	5.05
45 minutes	4.98	5.33	5.71	.....
60 minutes	6.18	5.69	5.89	5.88
70 minutes	6.22	.....	6.10	6.09
80 minutes	6.62	.....	6.37	6.41
90 minutes	6.22	6.18	6.47	6.74

[a] Spaghetti produced from blends of Ardex 550 plus No. 1 Semolina.

would not be lost during canning operations, because macaroni products are normally cooked only 10 minutes or less before being canned. The fact that the protein is extracted into water and that there is an increased per cent of solids extracted into the cooking water could be important factors in the formulation of spaghetti and other types of sauces. These facts could also influence the organoleptic evaluation of the consistency and texture of the sauce. This would be particularly true in evaluating cold samples. In the canning industry and in institutional feeding there is a definite desire and a need for macaroni products that are resistant to overcooking. Resistance to overcooking and other characteristics, i.e., color, odor, flavor, appearance, and palatability, were rated by taste-panel evaluation. Taste-panel evaluation of canned spaghetti in tomato-cheese sauce during six months at the Metal Division Research and Development Laboratory of Continental Can Co. (1959) showed that the product manufactured with Ardex 550 had significantly better firmness than that made from 100% semolina. The panel further concluded that No. 1 semolina plus 17% Ardex 550 was the most acceptable from the standpoint of flavor and texture and was satisfactory from the standpoint of firmness.

The addition of 12.5 or 17% Ardex 550 to semolina for the production of macaroni products substantially improved the relative protein efficiency. The use of 25% Ardex 550 with semolina improved the relative protein efficiency somewhat more, but not significantly. The quality of the protein as determined by rat assay by two laboratories illustrates the the importance of supplementation of semolina protein to obtain a nutritional product.

Fig. 6 shows comparative results for protein assay of semolina and semolina plus three levels of Ardex 550 that might be used for production of high-protein

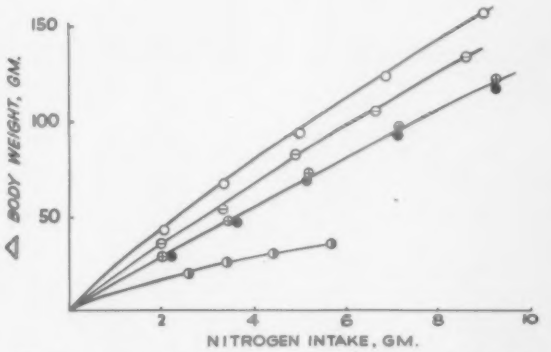

Fig. 6. A comparison of growth response of rats fed different types of protein for 42 days: ○ casein; ⊖ 25% Ardex 550 soy spaghetti; ⊕ 17% Ardex 550 soy spaghetti; ● 12.5% Ardex 550 soy spaghetti; ◑ semolina.

macaroni products. This graph demonstrates the improvement in the nutritional quality of semolina protein by the addition of soyflour. Allison *et al.* (1959) reported on nutritional studies with wheat germ, yeast, or combinations of wheat germ and yeast to supplement semolina. Their results confirm this nutritional improvement of semolina by certain protein supplementation.

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Freezer Burn as a Limiting Factor in the Storage of Animal Tissue. I.

Experiments with Livers Frozen Without Weight Loss

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(Manuscript received October, 1960)

SUMMARY

Slices from the surface of beef liver with varying fat content were frozen and subsequently exposed to slow air flow at −10° C and relative humidities of 78, 88 and 97% and the development of freezer-burn (fb) observed. The following effects were noted:—

(1) The maximum development was observed when samples from mature animals were rapidly frozen (4 min) without weight loss.

(2) No fb appeared on fetal livers, but the area and intensity of fb increased with increasing age of the animal from 2–3 weeks onwards.

(3) With livers from fully grown animals the onset of fb appeared at relatively low weight loss in storage, independent of the rate of evaporation during storage. The intensity of fb was independent of fat content but there were some differences between individual animals.

(4) Dehydration losses of up to 2.3 g/dm² while holding the samples at about 0° C before freezing caused a small but significant increase in the weight loss necessary to produce fb.

(5) A more pronounced delay was produced by the effects of incipient chemical changes in the liver when held at about 2° C for up to 7 days before freezing. This effect was less evident at high fat contents. There was a tendency for fb to occur at lower weight loss the lower the rate of evaporation in storage.

(6) As fb damage becomes more intense it becomes progressively more irreversible and while the early stages disappear on raising the relative humidity the most advanced stages are still evident after the samples are boiled.

Desiccation on the surface of frozen animal tissue sometimes produces a brightening of the color that changes with progressive storage from a whitish to an unsightly grayish-yellow appearance caused by an alteration of the tissue to a condition described as spongy or corky, with a honeycomb arrangement of air pockets that scatter incident light (Moran, 1934, 1937a, b). This desiccated appearance is designated as pock marking, scald spots, or freezer burn (fb).

Freezer burn should be distinguished from light and dark patches that result from varying ice crystal size when freezing rates vary, and also from the general decrease of opacity (Griffiths *et al.*, 1932) and darkening that occurs during surface dehydration as a result of concentration and oxidation of the pigments (Moran and Hale, 1929; Callow, 1952).

Present knowledge regarding the development of freezer burn. The majority of foodstuffs frozen in the original state, and also some frozen processed foods (Pearce and Reid, 1946), may develop fb. The presence of even the early stages of fb depreciates quality (Ziemba, 1946). Later stages of fb represent irreversible damage that remains after thawing but usually disappears after cooking (Snyder *et al.*, 1953) though it may cause off-flavors (Tressler, 1935) and denaturation of proteins. The increased effective area in tissue with fb favors oxidative rancidity of the fats (Cook, 1940), and the products of oxidation may accelerate the oxidation of the heme pigments (Watts, 1954). Fibers from muscles of perch (Partmann, 1955) and poultry (Partmann, 1959) affected by fb failed to contract on addition of adenosinetriphosphate, and in later stages the plasticizer effect ceased to appear. Pores found in such fibers and in the connective tissue were assumed to be pathways for the water vapor subliming from ice crystals. Freezer burn developed preferentially on the lean surface of rabbits (Vickery, 1953) and on the follicles of plucked poultry (Tressler, 1933).

Weight losses associated with the onset of fb varied greatly from several per cent (Tressler, 1935) to the very small losses in sealed containers due to sublimation-condensation effects caused by temperature fluctuations (Anon., 1933), and in the air pockets of flexible packages (Stewart, 1953). Wax-paper wraps with high permeability to water vapor and faulty heat seals (Bouton and Howard, 1956), and also temperature fluctuations (Hilder, 1946), favored the onset of fb on various meats. An increased rate of freezing did not retard the development of fb (Cook, 1940). No fb appeared at temperatures of −3.3° C to −6.7° C on cuts of meat of different types of animals, but it developed between −12° C and −29° C (Ramsbottom, 1947).

In experiments with poultry packed in airtight cellophane bags and uniformly frozen at temperatures between −10° C and −23° C, the extent of fb varied widely, and variation in fb was on the same order within experiments as between experiments (Snyder *et al.*, 1953).

Early attempts to control freezer burn. Loss of water vapor, the necessary antecedent to the onset of fb on frozen food, was restricted by using high relative humidities (98% at −14° C, 96% at −21° C) so that fb was prevented over a period of 6–10 months (Cook, 1940). Water-vaporproof containers (cans), boxes with linings, bags made of plastic films, aluminum foils, and laminates with adequate seals were recommended to serve the same purpose (Moran, 1934, 1937b; Tressler, 1935; Margolf *et al.*, 1956). To avoid pockets, cans and boxes should be filled to capacity (Pennington, 1947). Application of wax coatings and "glazing" with sugar solutions are other treat-

ments that have been recommended. The permeability to water vapor determines the duration of the protection provided by packages. Meat cuts packed in Cry-O-Vac, Pliofilm, or Cellophane M.S.A.T. developed fb when stored for several years (Ramsbottom, 1947).

Barnicoat (1934) reported the reduction of fb on offals by bringing about surface desiccation before freezing. Bergh (1955) treated meat before freezing with solutions of higher alcohols, hexoses, etc., to prevent discoloration including that due to fb.

Rehydration of frozen steaks in water removed patches of fb, but they were of less acceptable flavor than control pieces (Anon., 1946).

Present approach to prevention of freezer burn. An attempt has been made to obtain more precise information regarding the mechanism of fb development as a basis of treatments for its better control. It was first necessary to determine the conditions required to produce maximum fb.

The present paper deals only with experiments in which the freezing of samples was carried out without concurrent weight losses and without pretreatment of the surface. The effects of weight loss during storage, of weight losses prior to freezing and the duration of prestorage at chilled temperature, and of the composition of the material were studied.

The influence of weight loss during freezing and of the effects of surface treatments will be reported later.

EXPERIMENTAL

Freezer burn frequently develops in patches on the surface of a product of apparently uniform composition, and the differences in the characteristic properties of the frozen material that are responsible for the uneven development of fb are not known. To study the development of fb, beef liver was used since it provided a material from which surface samples of reasonable uniformity of composition and structure could be prepared.

The rate of evaporation from the surface of biological material cannot be kept constant, because the coefficient of evaporation decreases with time; but the drying power of the air can be controlled through control of temperature, humidity, and air flow. The equipment described below was designed with this requirement in view, and its performance was tested by determination of the rates of sublimation from the surface of standard ice samples.

Temperature control. Three experimental drums (Fig. 1) were arranged in a cabinet that was placed in a commercial cold store maintained at a temperature of −14° C. Fluctuations in temperature within the drums, and differences between the drums, did not exceed 0.2° C.

Control of relative humidity with solutions of known vapor pressure. The relative humidity in the drums was controlled by absorbing the water vapor evaporated from the sample, either with saturated salt solutions (with excess of salt) or with an H_2SO_4 solution, in plastic dishes at the bottom of the drums (Fig. 1). The values of RH assigned to these solutions (Table 1) were obtained from O'Brien's (1948) survey of RH of saturated solutions at temperatures above 0° C with extrapolation to lower temperatures by means of Othmer's (1940) relation between vapor pressure of solution and water at the same temperature, assuming insignificant change in concentration. The data in Table 1 were found to be in reasonable agreement with results from determinations of isopiestic solutions of H_2SO_4 using the RH concentration data of Collins (1933) extrapolated to −10° C.

Change of relative humidity due to evaporation from samples. The extent to which the relative humidity of the air will depart from that of the solutions, and the change in passing over the samples, will be determined by the rate of absorption in the solutions and the rate of evaporation from the samples, and these in turn will be determined by the coefficients of absorption and evaporation. The coefficients were determined as outlined in the appendix, and the dimensions of the equipment selected accordingly. The maximum changes in relative humidity to be expected in each drum with the maximum rates of weight loss found with the liver samples are given in Table 2.

Air flow. The speed of the air flow in the annular space in each drum was measured with a hot-wire anemometer calibrated in a wind tunnel under similar conditions of exposure. The

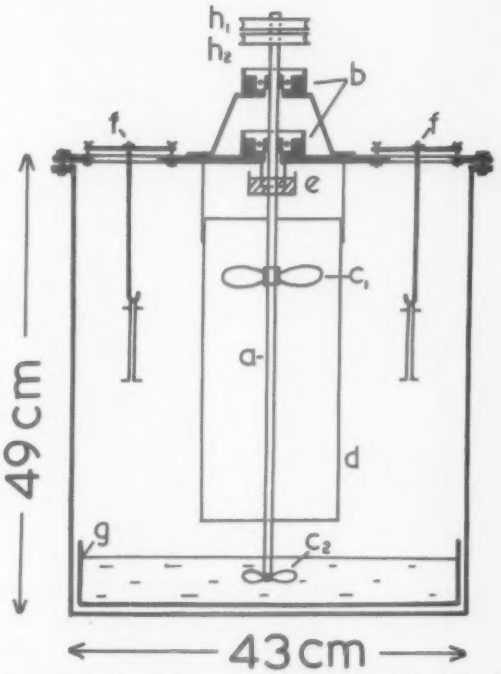

Fig. 1. Experimental vessel, with stainless-steel shaft (a) held in position by ball bearings (b) and operating propeller (c_1) for circulating the air and (c_2) for stirring the solution in the plastic dish (g); guide (d) for directing the air; oil seal (e); lids (f) with hooks for samples; and pulleys (h_1 and h_2) to drive the propellers and transfer the drive.

Table 1. Equilibrium per cent relative humidity of solutions used for control

Solution	Drum	Relative humidity at −10° C	
		Over water	Over ice
$Ca(NO_3)_2$ (sat.)	A	71	78
NaCl (sat.)	B	80	88
H_2SO_4 (4.6*N*)	C	88	97

Table 2. Departure of vapor pressure of air from equilibrium conditions

Drum	Air speed (u) cm/sec	Maximum rate of weight loss (W) g/sec × 10^6	Increase in vapor pressure on passing over samples $\left(\frac{100\,\Delta P_s}{P_1}\right)$ %	Increase of vapor pressure of air leaving solution over equilibrium pressure ($\Delta\Phi$) %
A	4.3	64.8	0.5	2.3
B	2.9	37.1	0.4	1.2
C	2.4	8.3	0.2	0.2

average speed for each drum is given in Table 2. Variations in speed from position to position within the drums were on the order of 13%, but were consistent from trial to trial.

Drying power. The drying power of the air was determined at four positions in each drum by measuring the rate of evaporation from cylindrical blocks of ice. The results were brought to a common basis by expressing the rates as grams per unit area per day and per unit vapor pressure deficit. The results of four replicate determinations are illustrated in Fig. 2, which shows the influence of air speed. The relation of these results to data by other workers is discussed in the appendix. An analysis of variance showed that the variations at any one position were larger than could be expected from errors in weighing. Since there were no consistent variations from trial to trial the variations presumably arose from random fluctuations in air flow rather than from other changes in the environment. However, such variations are small compared with those involved in making subjective estimates of the degree of fb.

Preparation of samples. Samples taken from the surface of beef livers were frozen and stored in stainless-steel trays 5 × 8 cm × 0.3 cm deep. A mechanical cutter (Fig. 3) was devised

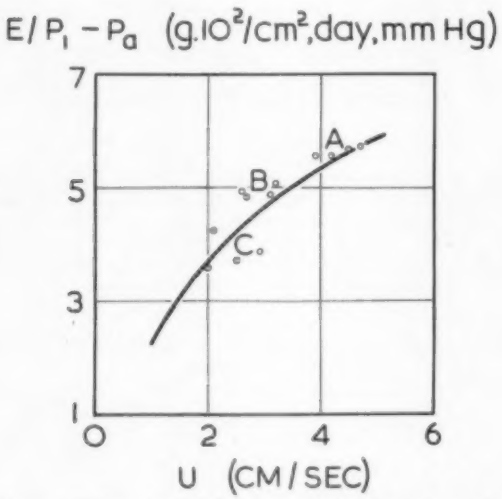

Fig. 2. Drying power of the air at different rates of air flow in the drums A, B, C as determined by evaporation from ice blocks at −10° C.

to prepare slices of uniform thickness. The cutting action is produced by the razor held in a shaft that reciprocates as a result of the eccentricity (2 mm) of the cam on the end of the shaft of the electric motor (1400 rpm). Contact between cam and shaft is maintained by the spring. Strips from the surface of the liver of the desired width are passed by hand under the moving blade. The distance between the knife and the bottom of the slide way can be set to give the desired thickness of the slices. The slices possess an even cut surface, and the accuracy of the thickness can easily be kept to a small fraction of 1 mm.

Samples cut to size (av wt 12.2 g) were normally put in the trays with the membrane uppermost, covered with a film of polyethene (slightly larger than the tray) to prevent evaporation, and then cooled to 0° C and frozen. When fb was studied on the cut surface, this surface was uppermost in the trays.

Chemical determinations. Moisture contents were determined by dehydration for 17 hours at 104° C of 5-g samples of the liver surface, mixed with 20 g sand. Fat contents were determined by Soxhlet extraction with ethyl ether for 4½ hr, and drying the extracted material for 1 hr at 104° C. The moisture content (% fat-free wet basis) and the fat content (% wet basis) of samples of the surface layer taken from the thick, middle, and thin portions of individual livers varied with

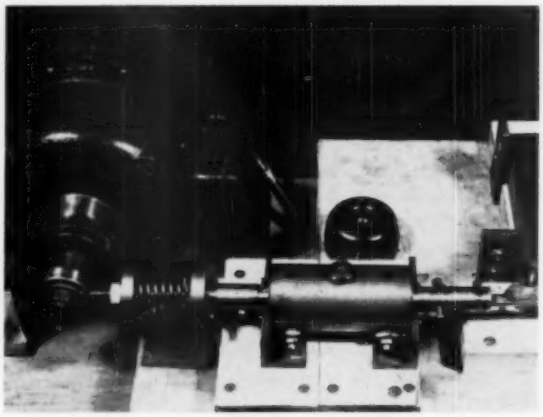

Fig. 3. Mechanical equipment for cutting liver slices.

position ($P<0.01$), both increasing to some extent from the thick to the thin end of the liver. Samples used to characterize individual livers were therefore composite samples of the whole surface.

Numerous studies have shown that in most animal tissue there is a correlation between fat and moisture. Fig. 4 shows, for all the livers used in this study, the relation between moisture content (fat-free basis) and fat content (wet basis). The distribution of moisture content at a given fat content was found to be non-normal, and therefore the Kruskal-Wallis (1952) non-parametric one-way analysis of variance was used to study the variation of moisture content among livers of mature animals, and it was found to be fat-dependent ($P<0.001$). Although a linear relationship would appear to fit the data reasonably well, seasonal and other factors are obviously operating. Low moisture content in liver can be expected during periods of drought and also after periods of starvation and transport (Anon., 1958). Moisture contents of fetal and calf livers were

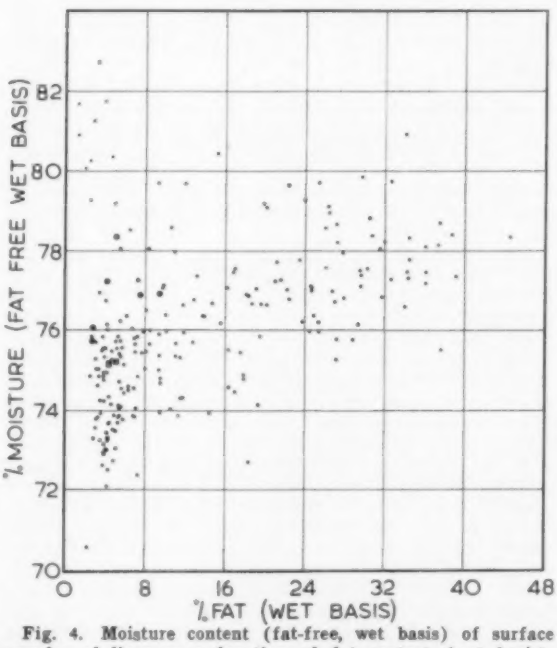

Fig. 4. Moisture content (fat-free, wet basis) of surface samples of liver as a function of fat content (wet basis). X, fetal livers; ●, livers from mature animals; ⊕, livers from calves; ⊠, livers from yearlings.

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significantly different ($P<0.001$) from livers of similar fat contents from normal adult animals. The two values for livers from yearlings were just significantly different from those of adult animals ($P<0.05$).

Freezing of samples. Rate of freezing was determined as the time necessary to decrease the temperature from 0° C to −10° C when 4 trays containing liver samples were floated on mercury at −21° C in a porcelain dish in a freezer room at −12° C. With this technique, which was used in all experiments reported in this paper, the freezing time, determined as the mean of 8 measurements, was 4.1 ± 0.6 min.

Estimation of the development of freezer burn. The development of fb was observed visually, and weight loss in storage was determined when the intensity reached three stages (Fig. 5):

I_1 Initial stage of fb with pin points and irregular small patches or streaks, but without development of light color.

I_2 Desiccation readily visible, with pin points and spots increased to about 2 mm diameter, and color approaching white.

I_3 Desiccation strongly marked. The area has ceased to increase, the white color tends to fade, and the surface tends to become rough.

Fig. 5. Liver slices with freezer burn of different intensities: top right, fresh sample; top left, freezer burn intensity I_3; bottom left, freezer burn intensity I_2; bottom right, freezer burn intensity I_1.

Liver samples with fb of intensities I_2 and I_3 were placed in Petri dishes with water-vaporproof seals at −10° C, and were sufficiently stable to be used as standards. Freezer burn of intensity I_1 is, to a large extent, reversible, and disappears after some time from samples held in sealed containers.

RESULTS AND DISCUSSION

The conditions for the maximum development of freezer burn on beef livers have been shown to be:

a) Livers must be taken from adult animals and used immediately after slaughter.

b) The liver slices must be covered with a plastic film that is practically impermeable to water vapor so that weight loss during freezing is insignificant.

c) The slices must be frozen quickly. A rate of freezing of 4.1 min for the slices used in this work proved sufficiently fast.

Freezer burn developed regularly on all samples treated according to this procedure, and was independent of the composition of the liver.

Effect of drying power of the air. Fig. 6 shows the increase of weight loss with time from samples with

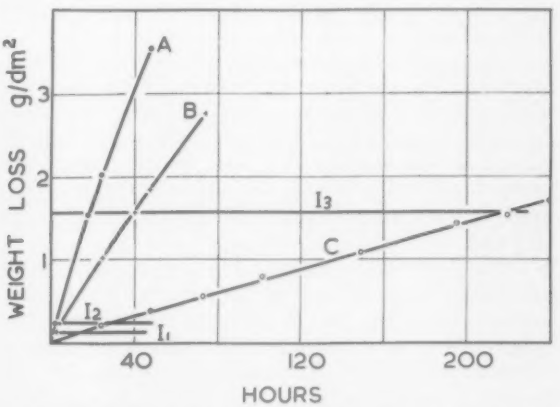

Fig. 6. Rate of weight loss from liver slices (5% fat) exposed to relative humidities of 78% (A), 88% (B), and 97% (C) at −10° C.

5.0% fat. With small losses (drum C) the increase in weight loss with time was practically linear, but with larger losses there was a slight decrease in rate with increase in time. High fat content (>20%) reduced the rate of weight loss only slightly, and differences were noticeable only after longer storage times than those necessary to produce fb of intensity I_3.

Analysis of variance of the weight loss at a given intensity (using log weight to obtain a uniform error variance at all intensities) showed that under conditions of maximum development there were no differences between the drums or between positions within the drums.

The average absolute levels of weight loss necessary to produce fb under the most favorable conditions for development are given in Table 3.

Effect of age and condition of beast. The formation of fb differed markedly with the age of the animal (Table 4). Slices of liver from animals of different ages, frozen without weight loss in 4.1 min, were exposed to a relative humidity of 78% in drum A. Freezer burn was in no case observed on the surface of fetal livers (weight losses up to 8 g/dm²), and

Table 3. Weight loss necessary to produce freezer burn on liver slices under maximum conditions.

Conditions of exposure	Weight loss g/cm² at		
	I_1	I_2	I_3
Membrane exposed to evaporation	0.12	0.23	1.57
Cut surface exposed to evaporation (2.9% fat)	0.36	0.67	4.6
Cut surface exposed to evaporation (28.9% fat)	0.17	0.38	0.92

this was confirmed in further histological studies. Considerably higher weight losses during storage were needed to develop a given intensity of fb on livers of calves about 14 days old than on livers of older animals. Livers of fully grown animals required the smallest weight losses to produce fb of given intensity. Freezer burn proceeded to develop normally from intensity I_2 to more advanced stages on livers from all animals older than two weeks. The causes of the influence of the age of the animal on the formation of fb are not yet understood.

Exceptions to the normal development on livers of adult animals occurred only when animals were exposed to severe starvation, after extended periods of drought. The onset of fb was then postponed, and in a few cases did not appear at all. The increase of the intracellular osmotic concentration (Riecker *et al.*, 1957) of the liver cells during the period of stress may be a cause of the inhibition.

Effect of desiccation before freezing. Samples were taken from livers of varying fat content from mature animals and were frozen without weight loss after being held at 0° C in a dry atmosphere to produce varying weight losses. The results were compared with those for livers frozen without weight loss immediately after slaughter.

Livers (4.5–6.4 kg) have a ratio of area to weight of 7.6 dm²/kg that varies little over the above weight range. A weight loss of 1%, corresponding to a surface desiccation of 1.3 g/dm² (liver 5.9 kg), can be expected if livers are cooled without protection against evaporation before freezing. Average weight losses of 0.51, 1.04, and 2.30 g/dm² were applied. Freezing of the samples had to take place without delay to avoid equilibrium between the lower water content of the surface and the practically unchanged water content of the deeper layers.

The results in Fig. 7 (compare b with a) reveal that the weight loss required to produce fb on samples desiccated before freezing was significantly ($P<0.001$), but only slightly, greater than for the controls. An analysis of variance showed little evidence of a regression on fat content. There was some evidence that, especially at high fat content, fb developed at lower weight loss, the lower the rate of evaporation during storage.

Excessively high weight losses (8.4 g/dm²) before freezing, which also cause severe dark discoloration, were needed to prevent the appearance of fb on livers with normal to medium fat content. However, the samples had to be frozen shortly after the drying of the surface. If freezing was delayed for 16 hr at 0° C and further evaporation prevented by covering the slices with polyethylene film, fb of all intensities was produced during storage at −10° C. Obviously the surface regained enough water from deeper layers to make fb possible.

Effect of storage at 2° C before freezing. Studies were made on livers of varying fat content stored for 4 and 7 days at 2° C before freezing without weight loss. The results are in Fig. 7 (compare c and d with a).

The weight losses during storage required to produce fb of given intensity were higher ($P<0.001$) than with the controls. With the material stored for 7 days there was clear evidence that, at least at intensities I_1 and I_2, the necessary weight losses de-

Table 4. Weight loss necessary to produce freezer burn of intensity I_2 on livers from animals of various ages (Storage at −10° C over Ca $(NO_3)_2$ solution with 78% relative humidity)

Age of animal	No. of livers	Initial weight of livers (kg)	Weight loss (g/dm²)
Fetal	3	0.28–0.40	no fb at 6.6
Fetal	3	0.40	no fb at 8.2
Approx. 2 wk.	2	0.62–0.84	0.62
18 months	2	2.3 & 2.4	0.29
Approx. 4 years	1	4.3	0.25

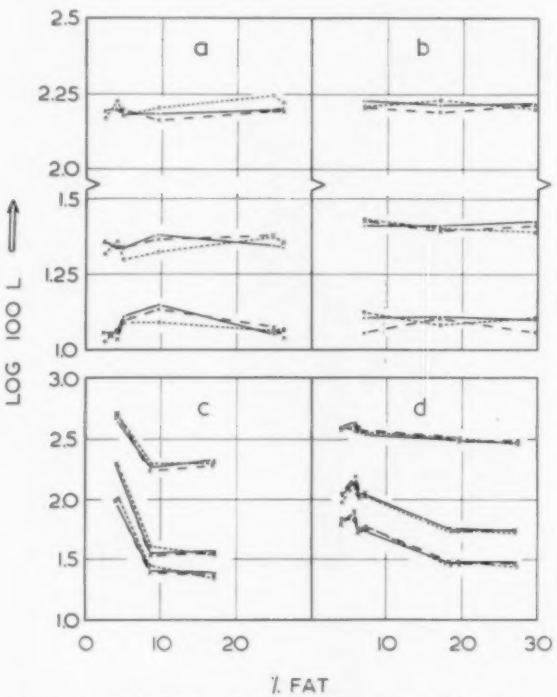

Fig. 7. Weight loss (g/dm²) necessary to produce freezer burn of intensities I_1, I_2, and I_3 on liver surfaces exposed to relative humidities of 78% (A .——.), 88% (B x—x), and 97% (C o....o) at −10° C with tissues of various fat contents. a) control; b) desiccated before freezing; c) held 4 days; and d) held 7 days at 2° C before freezing.

creased with increase of fat content. With the material stored for 4 days a similar trend was evident but the effect could also be explained by individual carcass differences. With the material stored 7 days there was also evidence that the relationship with fat content is more evident, the lower the rate of evaporation in storage.

Paper chromatographic studies have shown that during the 7 days' storage at 2° C an increasing concentration of amino acids occurs in extracts of the livers, indicating a breakdown of peptides or more complex nitrogenous compounds. This could be partly due to bacterial activity, but an increase of the amino acid concentration due to proteolytic activity alone was observed in liver by Zender *et al.* (1958). It is probable that such chemical changes during storage have an inhibiting effect on fb.

Freezer burn development on cut surfaces. Liver samples were prepared and frozen without weight loss, but the cut surfaces were exposed to desiccation in the drums.

Freezer burn appeared on the cut surfaces, but its characteristic appearance on a lean liver slice (3% fat) was less bright than fb on the membrane surface. The weight losses required to produce the three stages of fb on cut liver surfaces were higher than with membrane surfaces. Average values are given in Table 3 for comparison with membrane surfaces.

With fat samples, fb of the cut surface more closely resembled the typical fb on membrane surfaces.

Freezer burn can also be produced on Glisson's capsule alone after its removal from the liver. Samples (5 × 8 cm, 0.6–0.8 g) were frozen without losses. The membrane was generally too thin to allow the development of intensity I_3. Prefreezing losses at 0° C had no noticeable effect on the development of fb on the membrane.

Reversibility of freezer burn. Water vapor condensed on the surface of a frozen liver with fb forms an ice layer without filling the holes and capillaries of the dehydrated tissue. During thawing, however, water may penetrate the spaces and capillaries, and the original bloom may be wholly or partly restored. The normal appearance can be restored if the desiccation losses during storage are not greater than 1.7 to 2.1 g/dm^2. However, if the fb is allowed to develop to an advanced stage so that the weight loss exceeds 6 g/dm^2, the desiccated appearance will still be noticeable after the slices are boiled.

The vapor pressure over the frozen liver samples, which remains constant as long as ice crystals are present at the surface, is lowered and tends to approach the partial pressure of the air after the sublimation of the ice, the cells near the surface being closest to equilibrium. An RH of 97% (rel. to ice) was sufficient to produce fb of intensity I_2, which cannot return to the original condition when the sample is stored in an atmosphere practically saturated with water vapor at −10° C. The state of desiccation represented by freezer burn of intensity I_3 is obviously irreversible.

Theoretically, when the tissue is depleted of ice crystals and additional water has been evaporated to establish fb at an equilibrium humidity (rel. to ice at −10° C) of, for instance, 88%, the same equilibrium concentration of solutes in the tissue should be established as can also be obtained by freezing liver without weight loss to a lower temperature, in this case −21° C. It is obvious, however, from general experience that liver frozen at −21° C without weight loss exhibits, neither in the frozen nor in the defrosted state, damage similar to fb at −10° C.

The changes caused by freezing are therefore slight, and irreversible changes associated with fb are evidently due mainly to dehydration. The production of air-tissue interfaces is probably an important factor. It is to be noted, however, that in freeze-drying, where the extent of dehydration is high but oxygen is practically eliminated during processing, though there is some evidence of irreversibility, the undesirable changes are substantially reduced. The rate of the dehydration process is probably important.

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APPENDIX

Determination of coefficients of evaporation and absorption and maximum variation of relative humidity. In the closed system within each drum a state of equilibrium will be set up between the evaporation of W g ice per sec from n samples with vapor pressure P_1 mm Hg and its absorption into the salt solution with a vapor pressure of P_2 mm Hg and at the same time the vapor pressure of the air will rise from P_a to P_a' as it passes the samples.

At equilibrium $W = nk_1(P_1 - P_a) = k_2(P_a' - P_2)$ where k_1g/sec/mm Hg is the coefficient of evaporation for a given size of sample of ice and k_2 g/sec/mm Hg is the coefficient of absorption for a surface of solution of area equal to the plastic dish.

The air is driven by the fan through the annular space of area a cm² at a velocity of u cm/sec. After having passed over the samples and taken up W g/sec of water vapor, its vapor pressure will rise by ΔP_a where $\Delta P\hat{a} = P\hat{a}' - P_a =$

$$\frac{W \times 22.4 \times 760 \times 263 \times 10^3}{18 \times u \times a \times 273} = \frac{9.11\ W \times 10^5}{u\ a}$$

mm Hg at a temperature of −10° C.

The relation between k_1 and k_2 is then given in terms of measurable or known quantities by

$$\frac{1}{k_1} = n\left[\frac{P_1 - P_2 + \Delta P_a}{W} - \frac{1}{k_2}\right]$$

and the values of k_1 and k_2 can be determined by measuring W for two different values of n.

From preliminary measurements of k_1 and k_2 made on a model, the dimensions of the experimental drums were selected. The coefficients were then redetermined using data for rates of loss from 2 and 4 cylinders of ice each 4 cm in diameter and weighing 75 g. For the drum with saturated $Ca(NO_3)_2$ solution the coefficients were $k_1 = 0.73 \times 10^{-4}$ and k_2 1.2×10^{-4} g/sec/mm Hg (u = 4.3 cm/sec, a = 1385 cm², $P_1 = 1.95$ mm Hg, $P_2 = 1.50$ mm Hg, area of solution 1390 cm², temperature −10° C).

The relative humidity of the air that has passed over the solution exceeds the relative humidity of the solution by $\Delta\Phi$ where $\Delta\Phi = \frac{P_a - P_2}{P_1} \cdot 100 = \left[\frac{W}{k_2} - \Delta P_a\right] \cdot \frac{100}{P_1}$ and the rise in relative humidity on passing the samples is given by $\frac{\Delta P_a}{P_1} \cdot 100$. These are thus expressible in terms of W and k_2 and their maximum values for rates of evaporation found in the experiments are calculable as in Table 2.

Effect of velocity on drying power of air. Powell (1940) has shown that the rate of evaporation into a rapid air stream from a surface of given geometrical shape can be represented as a single curve by plotting dimensionless groups. With cylinders of constant size and with constant air temperature the relationship can be expressed as that between $E/(P_1 - P_a)$ and u where E is the rate of evaporation per unit area (g/sec/cm²), P_1 and P_a the water vapor pressure (g/cm²) at the evaporating surface and in the air stream respectively, and u the rate of air flow (cm/sec).

Powell, using velocities above 50 cm/sec, obtained an approximately exponential relationship. In the present work much lower velocities were used and the experimental conditions necessary for the freezer-burn studies were difficult to reconcile with equilibration within the air stream. It is not surprising therefore that the data (Fig. 2) do not fit Powell's relation. The arrangement, however, appears to give consistent results over long periods, and the data can be fitted to the formula

$$\frac{E}{P_1 - P_a} = 4.30 \times 10^{-5}(1 - e^{-0.22u}) + 0.48 \times 10^{-5}\ u^{0.46}$$

which satisfies the physical fact of zero evaporation at zero velocity and restricted convection and, at higher velocities when the second term predominates, gives an exponent that corresponds closely with the value 0.8 found by Powell for evaporation from circular cylinders with parallel air flow at higher speeds.

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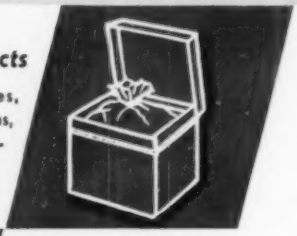

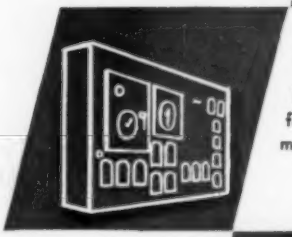

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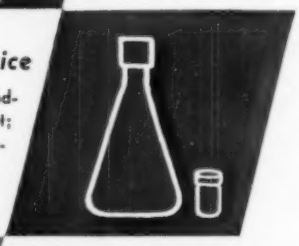

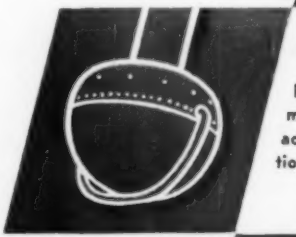

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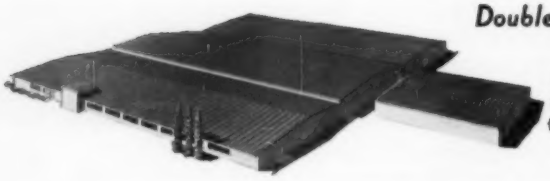

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Freezer Burn as a Limiting Factor in the Storage of Animal Tissue. II.

Histological Study of Freezer Burn of Liver

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(Manuscript received October, 1960)

SUMMARY

Sections from samples of liver fixed in the frozen state showed that dehydration of the surface at −10° C first brings about the formation of a condensed layer, and then, from this layer freezer burn (fb) develops, beginning near the surface. As dehydration continues, development of fb cavities spreads to greater depths of the condensed layer, thereby increasing the intensity of the fb. This development tends to be greatest along the vessels in the tissue and along fibers in the capsule. At the same time the sharply defined lower boundary of the condensed layer advances toward the interior, where the sublimation of the ice crystals causes the shrinkage of further tissue and the condensed layer increases in thickness. In fetal tissue, a condensed layer only is formed, without the typical porous structure, and the liver shows a dark discoloration but not fb.

Liver slices were frozen rapidly without weight loss and stored at −10° C and 78% relative humidity to produce freezer burn (fb) as already described by Kaess (1961). Samples both before and immediately after freezing, and samples with fb of intensity I_2 and later stages were used for histological examination. Spots and streaks of fb of intensity I_1 were sparsely distributed and located only on the top layer.

SAMPLES FIXED AT ROOM TEMPERATURE

The slices were fixed in neutral 10% solutions of formalin. Nuclei and cytoplasm were stained with hematoxylin-eosin stain, and connective tissue by the hydrochloric acid–methyl blue method of Lillie (1952).

Formation of cavities from desiccation. Freezer burn affected first the top layer of Glisson's capsule, where optically empty spaces appeared in sections from samples with fb of intensity I_1, which indicated holes caused by desiccation (compare Fig. 1-a,b). Also, the squamous epithelium was ruptured in a number of places. Individual fibers were easily discernible in the unaffected part of the capsule.

When fb advanced to intensity I_2 (Fig. 1-c), the frequency of the holes in the top layer of the capsule did not appear to increase, but the subsequent layer, marked by its large content of coarse collagen fibers, had developed large numbers of holes uniformly over the section. The affected area extended from the capsule into the liver cells. With further desiccation the front of the holes penetrated to deeper layers of the liver cells, and Fig. 1-d shows a stage of fb that conforms closely to Moran and Wright's (1937) photograph of fb on kidney.

Advanced fb hardly affected the collagenous and elastic fibers of the top layer of the capsule, but the coarse collagen bundles seemed to split or to be on the point of splitting where holes had formed. Between holes, the bundles became more compressed with increasing weight losses, the outlines of single fibers disappeared, and the nuclei became shriveled.

The liver cells below the capsule in the immediate vicinity of the freezer-burn holes, and also the Kupffer cells, became wrinkled with nuclei of reduced size, and were even disintegrated when the desiccation reached an advanced stage, especially in fat livers. With increasing weight losses the remaining patches of cells around the fb holes assumed a more and more condensed structure, in which it was difficult to discern details.

In some cells of lean livers, sickle-shaped splits appeared and increased in number and size as dehydration proceeded until the splits in neighboring cells finally widened to form one cavity. Fat tissue did not show such splits. The cavities produced during the development of fb tended to follow the course of the vessels, especially the sinusoids. In later stages, cross connections developed between them. In the capsule the fb holes were arranged in the direction of the fibers.

In sections from slices of lean livers that developed fb when the cut surface (capsule absent) was exposed to desiccation, the holes seemed to be more sparsely distributed (Fig. 2).

Effect of fat content. In livers of low and normal fat content (not greater than 10%), the groups of holes found close to the outer surface of the capsule were more sparsely distributed in samples with fb of intensity I_2 than in fat livers, and the adjoining layer of the capsule with the accumulation of coarse collagenous fibers was generally not affected at all. The holes produced between the liver cells showed more ragged outlines than in fat livers, and high magnification revealed fragments of cytoplasmic origin in the fb spaces. The depth of penetration generally seemed to be less in lean livers than in fat livers for weight losses of the same order.

In lean livers Jackson's staining technique (Jackson, 1944) for lipids showed single well-defined globules in the liver cells. With increasing fat content (15% and greater), first the size and then the number of globules increased, and finally, starting in the portal area, the single globules merged into one big globule. In tissue damaged by fb the fat globules of adjoining cells merged into irregular masses.

SAMPLES FIXED IN THE FROZEN STATE

Since sections from frozen samples fixed at room temperature failed to show the position and the shape of the ice crystals in the tissue, samples were also fixed in neutral 10% formaldehyde solution kept in a constant-temperature bath at its freezing point (−2.9° C) so that small amounts of ice crystals were always present. This method prevented melting of ice crystals still present in the sample at −2.9° C. Some growth of the ice crystals in the sample may have occurred owing to the higher temperature of the freezing point, compared with the original temperature of the sample of −10° C. In later experiments neutral solutions of formalin were used at a strength such that the freezing point was close to the storage temperature of the samples.

Ice crystal formation. Standard samples 3 mm thick were frozen in 4 min, that is, at an approximate rate of 0.8 mm/min. The actual rate of crystal formations, which could be observed propagating along the vessels, was up to 4 times as great as this. Most of the ice appeared as large crystals in the extracellular spaces of the capsule and between the liver cells, but a number of small crystals were formed intracellularly also, as can be seen in the sample freeze-fixed immediately after freezing (Fig. 3-a). During storage at −10° C and 78% relative

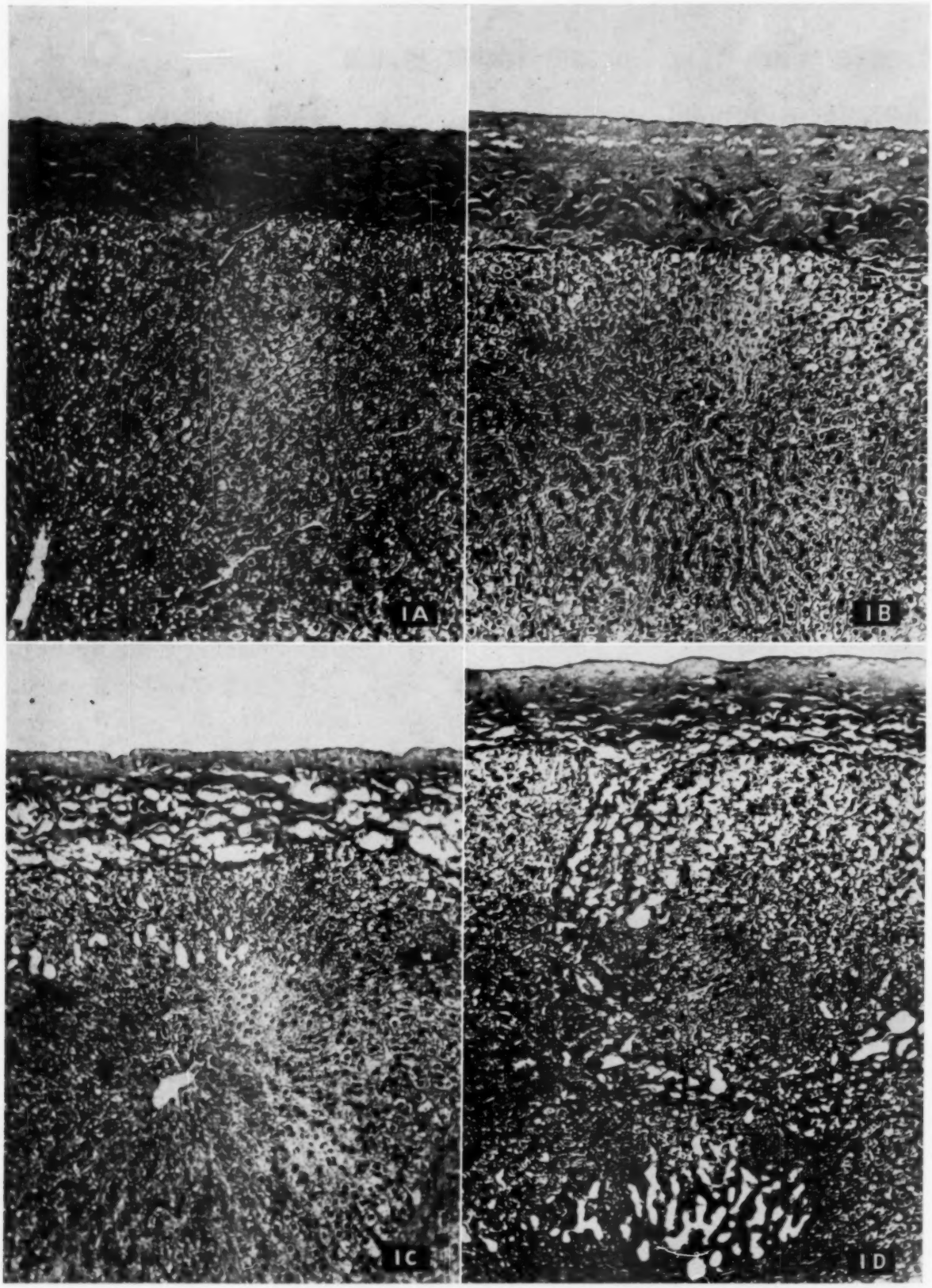

Fig. 1. Sections from liver slices (20% fat) frozen in 4 min without weight loss, fixed in neutral 10% formalin at 25°C, hematoxylin-eosin stain. ×445. a) immediately after freezing; b) freezer burn of intensity I_2; c) freezer-burn of intensity I_3; d) advanced freezer burn with weight loss of 4.0 g/dm.[2]

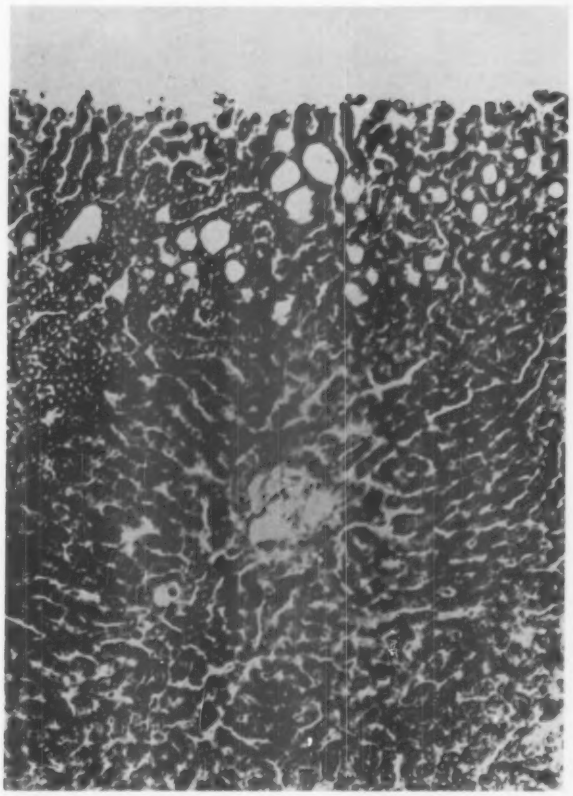

Fig. 2. Freezer burn of Intensity I_3 on the cut surface of liver slices (10% fat), fixed as in 1, stained with Jackson's acetic-carbol-sudan method. ×451.

humidity the smaller crystals seemed to vanish, probably by sublimation into larger forms.

Development of freezer-burn cavities. It appeared that after the sublimation of the ice, the fibers and liver cells merged into a dense layer of tissue, and it was only when additional evaporation of water took place from the concentrated salt solution and from the tissue that the formation of the cavities typical of freezer burn occurred. In samples with developing fb (intensity I_2) the condensed layer was still thin. In liver with fb of intensity I_3 the capsule and a small part of an adjacent layer of tissue showed an abundance of holes produced from a formerly condensed layer (Fig. 3-b). The condensed layer immediately below had a clearly defined boundary with the underlying region containing the ice crystals, which maintained a uniform size. As desiccation continued, the lower front of the condensed layer advanced to greater depths of the tissue, and on the opposite side the layer with fb gained in thickness. In consequence, the cell residues in the fb layer shrank further by drying out, starting from the top layer. The rate at which the condensed layer opened up was apparently slowed down as it increased in thickness. Fig. 3-c shows, for an advanced stage, the layer with fb, the condensed layer, and the remaining tissue with the spaces formerly filled with ice crystals. In fat liver tissue the condensed layer was less regularly formed and was thinner or even interrupted in the portal areas, where the fat is deposited more densely. Wang and co-workers (1954), working with muscle tissue (biceps femoris, semimembranosus) frozen at −80° or −150° C and subsequently freeze-dried, observed a narrow transitional zone of densely arrayed fibers between the frozen core and the lyophilized outer zone of the sample.

The appearance of the surface of the tissue with fb of intensity I_3 changed little as dehydration continued. The histological sections showed, however, that the damage continued to advance to considerably deeper layers.

The tissue of fetal livers in no case developed fb, even when weight losses were large. Fig. 4-b shows a section from a sample of fetal liver with extensive dehydration. No holes typical of fb could be detected in the large condensed layer, and split cells were absent.

Influence of treatments before freezing. No differences in microscopic appearances were detectable between sections from samples with f, that were dehydrated up to 2.9 g/dm² at 0° C before freezing and those from untreated controls.

Sections from liver slices stored for 7 days at about 2° C before freezing showed fewer holes in the condensed layer, and the spaces formerly occupied by ice crystals seemed to be smaller than in sections from the controls. Sections from the same livers fixed at room temperature showed large amounts of cytoplasmic substance (stained with eosin) in the fb holes. In the liver cells between holes the nuclei appeared to be shrunken, the cell walls disrupted, and the outlines of the cells faded.

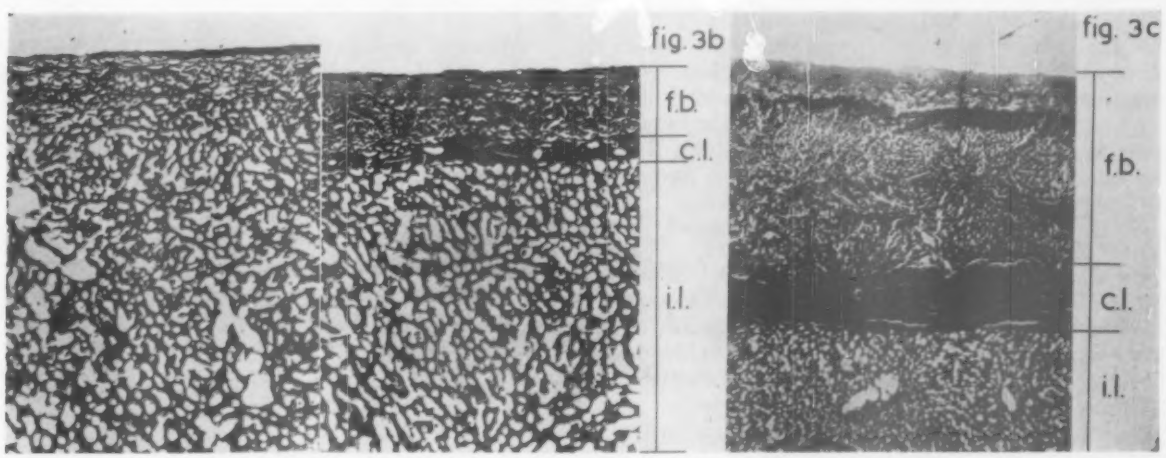

Fig. 3. Sections from liver slices (4.7% fat), freeze-fixed in neutral 10% formalin at −2.9°C, hematoxylin-eosin stain. a) immediately after freezing (×252); b) freezer burn of intensity I_3 (×252); c) advanced stage of freezer burn with weight loss of 9.4 g/dm² (×125). fb = layer with freezer burn; cl = condensed layer; il = layer with spaces previously occupied by ice crystals.

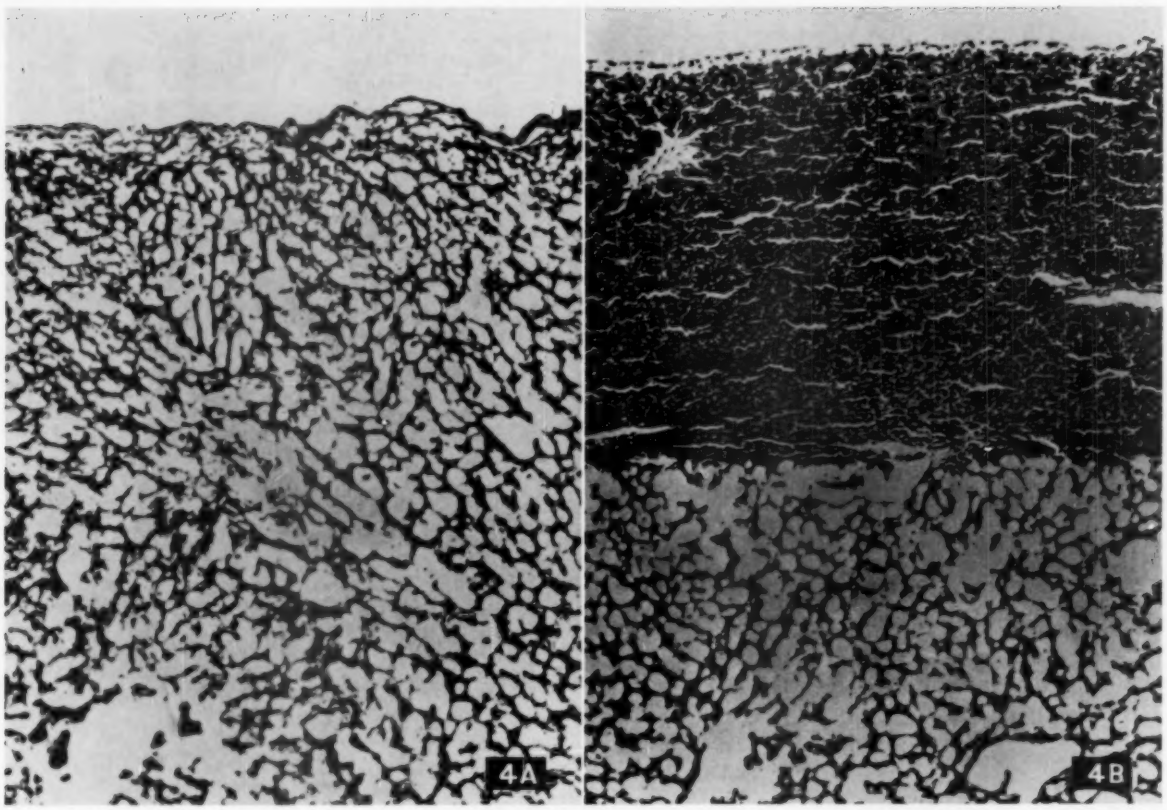

Fig. 4. Fetal liver, freeze-fixed as in 3, hematoxylin-eosin stain. (× 445). a) immediately after freezing; b) sample with 9.5 g/dm² weight loss.

DISCUSSION

Freeze-fixed sections from samples of frozen liver showed clearly that sublimation of the ice crystals does not, as might be assumed, give rise directly to the cavities in the tissue typical of fb, but first causes the formation of a condensed layer of tissue. It depends then on the nature of the tissue whether further weight losses result only in a growth in depth of the condensed layer, or whether, with continued desiccation, cavities are initiated in the upper part of the condensed layer while further sublimation of ice produces an advance of its lower boundary. In the first case the color of the surface darkens, and in the latter case the holes scatter the incident light, and the brighter discoloration of fb appears.

The texture and composition of pre-natal liver tissue exclusively favored the formation and growth of a condensed layer with subsequent dark discoloration. With advancing age of the liver, fb appeared to an increasing extent, together with patches of condensed areas, and livers of normally fed mature animals produced the maximum amount of fb during storage, when rapid freezing without weight loss was applied. High fat content increased the extent of fb, and the initiation of chemical changes by storage before freezing retarded the onset of fb.

It seems that conditions in the tissue that bring about increased cohesion or viscosity within the protein system and that produce a viscid state while desiccation proceeds, cause the tissue to move in the space left by subliming ice crystals and favor the formation of a condensed layer. On the other hand, conditions (such as fat accumulation and increasing age) that evidently produce a looser combination of the constituents of the tissue, promote the appearance of fb.

Obviously any loss of moisture from the condensed layer, as evaporation continues from the concentrated solution of tissue salts and from the condensed fibers, not only can give rise to fb but will also promote irreversible changes in the tissue. Partmann (1955) showed that the enzyme system of fish muscle is increasingly damaged as fb gains in intensity.

When typical fb cavities form in the condensed layers after sublimation of the ice crystals, irreversible damage of the frozen samples begins to develop and is favored by the presence of the concentrated solution of tissue salts. The increased effective surface introduced by the cavities may also cause additional irreversible oxidative reactions in the presence of air.

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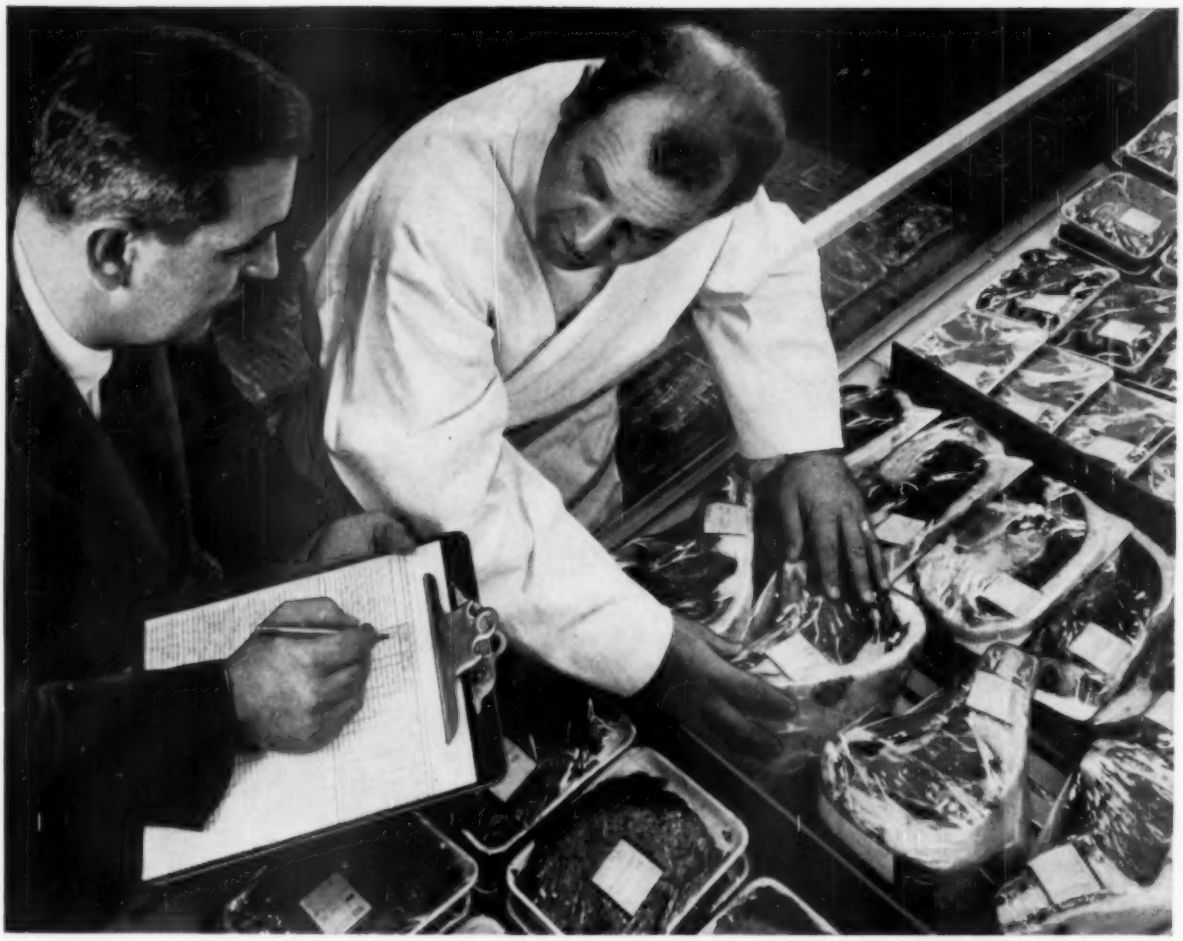

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The Effect of Oven Air Temperature, Circulation and Pressure on the Roasting of Top Rounds of Beef (Yield and Roasting Time)

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(Manuscript received May, 1960)

SUMMARY

Using a specially designed oven, the effects of temperature and air circulation on weight and volume yields of beef roasts were studied. It was found that the two yield values are directly related. Yield was found to be a function of evaporation loss and decreased as temperature and air circulation increased. However, at low temperature, with forced air circulation, and at the pressure of saturated steam (at oven temperature), yield increased and roasting time decreased.

INTRODUCTION

Heat within an oven is transferred to a product by a combination of conduction, convection, and radiation. The heat-transfer equations governing these processes indicate that the rate of heat transfer will depend primarily on temperature (differential); velocity of fluid; pressure (density) of fluid; and product (area, thickness). By careful product selection, area and thickness can be maintained nearly constant.

The effects of oven air temperature, circulation, pressure, and their interrelationships on the roasting of meat (yield and roasting time) were investigated to determine whether roasting losses and roasting time can be decreased simultaneously.

PROCEDURE

Design and construction of experimental TCP oven. Primary considerations in the design and construction of the experimental TCP oven (Fig. 1) (temperature, circulation and pressure) were the parameters supply, measurement, and control of oven air temperature, circulation, and pressure. Secondary considerations were the geometry of the test product, control oven volume (standard commercial electric oven of the type commonly used), standard pan sizes, and measurement of test product internal temperature.

The experimental TCP oven consisted of four independent but interrelated systems connected into an oven heating chamber—the oven air temperature, circulation, and pressure supply; measurement; and control systems; and the test product internal temperature measurement system.

All of the above systems affected the design and construction of each other and the oven heating chamber.

A review of the literature indicated that temperatures as high as 500° F and as low as 150° F could conceivably be used for roasting.

Temperature of the oven air was measured at 6 points, continuously, by iron-constantan thermocouples (20-gauge). These thermocouples were connected to a strip-chart potentiometer (Minneapolis-Honeywell Regulator Company). All thermocouples were shielded to reduce the influence of radiant heat on the thermocouples (Fig. 2).

It is generally agreed that forced convection will increase the rate of heat transfer. It was decided to use natural convection (present method of roasting) as the low limit of oven air circulation and the changing of the oven air every 6 sec as the high limit of oven air circulation. A shuntflo meter, model 3KS-9760, manufactured by Builders-Providence Company, was selected to measure air circulation in the TCP oven.

Steam pressure cooking is conducted at 5–15 psig. The pressure used in steam cooking is usually that of saturated steam at the temperature of cooking. No work has been reported on *air* pressure cooking (not saturated steam) of meat. Atmospheric pressure (present method of roasting) was used

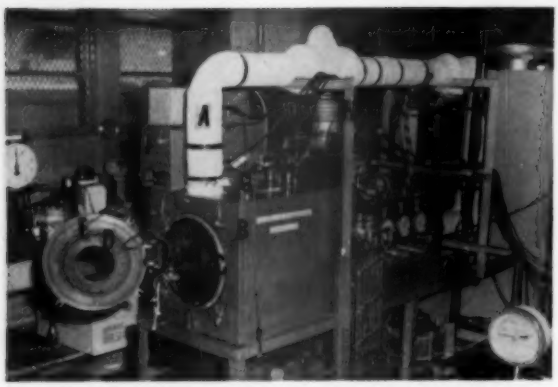

Fig. 1. View of experimental TCP oven showing (A) air duct, (B) oven chamber housing, (C) silicon door gasket, (D) oven-door sight glass, (E) oven door, (F) thermistor selector switch on electronic temperature control, (G) Conax thermocouple gland, and (H) air-duct support framing.

as the low limit of pressure, and 67 psia (pressure of saturated steam at 300° F) was used as the high limit of pressure.

The pressure of the oven-air in the TCP oven was measured by both recording and indicating gauges. The recording instrument was a Bristol model 29 pressure strip-chart recorder. Exact pressure indication was obtained from the Grove pneumatic gauge comparator.

Selection of control oven. A standard Navy type-60 electric roast oven (440 volts), procured under specification MIL-C-001218A, was selected for use as the control oven. This type of oven has 3 roasting compartments positioned vertically. Each oven compartment has a damper and an adjustable thermostat. This type of oven is widely used for baking and roasting. The oven was operated at control environment of 300° F, natural convection, and atmospheric pressure with the damper closed. Oven air temperature was measured at 9 points in the same manner as for the TCP oven. Natural convection and atmospheric pressure were not measured.

Variation. To minimize variation between and within animals, a 20-lb top round of beef, choice grade, was cut into 2 approximately equal halves. These paired rounds were stored and thawed together and used for the same experiment. One randomly selected half was roasted in the experimental TCP oven under experimental environments, and the other half was roasted in the control oven under control environment. The differences between the dependent variable measurement between the 2 halves should be caused only by the difference between the experimental oven environment and control oven environment plus experimental error. (Difference between and within rounds was minimized, not eliminated.)

The control top rounds of beef were always roasted under the control oven environmental conditions to an internal measured low-point temperature of 150° F. Since different top rounds of beef were used for each control test, the measured dependent variable among control tests should be due to the variation among rounds plus an experimental error. This variation among rounds obtained in the control oven under the environment and to an internal temperature (150° F medium-rare) recommended practically unanimously by cookbooks and commercial operators, was assumed to be the variation that will be found in the measured dependent variable in what is considered to be an acceptable product. A total of 40 paired experiments were conducted; 5 were replicated.

Test product. Paired top rounds of beef were removed from freezer storage (0° F) and thawed at 40° F for at least 84 hr before use. On removal from freezer storage, the product weight (oz) and volume (cc) were determined. After thawing, the product thawing time (hr), weight (oz), and volume (cc) were determined and recorded. The tare weight (oz) of the oven pan and rack was then determined. The test product was then put on the rack, with thermocouple multiple meat probes (Fig. 3) inserted, and placed into the hot oven. The test product was considered roasted and was removed from the oven when the internal measured low-point temperature reached 150° F.

Oven environment and variable measurement. Both the control and experimental ovens were stabilized at test conditions before the test product was placed in the oven. The temperatures of both ovens were continuously recorded, beginning about 2 hr before the start of the test and ending on removal of the test product from the oven.

The center compartment of the control oven was used for the test. The temperature of the center compartment was maintained at an average of 300° F. All compartments were operated with damper closed and heating elements on high heat. The oven door remained closed throughout the test.

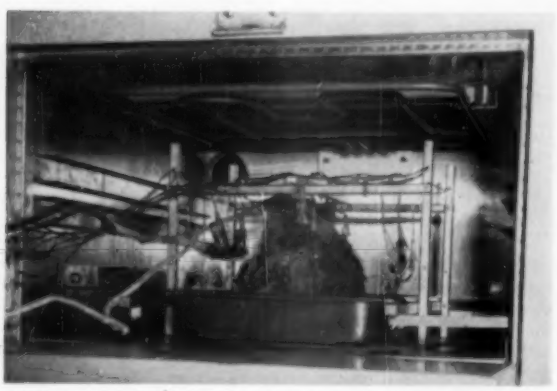

Fig. 2. Test product is shown in the control oven. The aluminum frame was constructed to hold the thermocouples in place. The thermocouples are shielded to reduce the influence of radiant heat on the thermocouples so that true oven air temperature will be measured.

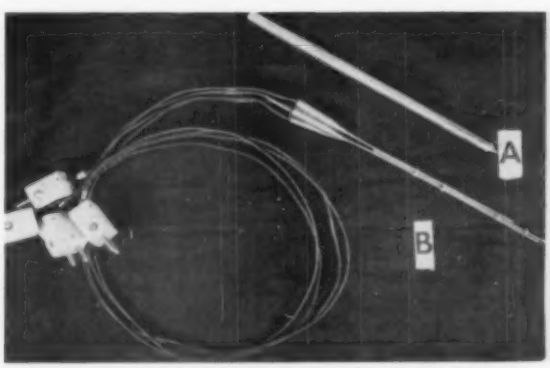

Fig. 3. The thermocouple multiple meat probe consists of 2 basic parts—the temperature probe and its protecting jacket. Two probe types were constructed, one a 5-point probe and the other a 4-point probe. These probes differed in construction only in the number of thermocouple points. Each probe consisted of iron-constantan 22-gauge thermocouples, packed in magnesium oxide under pressure into a stainless-steel tube 1/16 in. O.D. The thermocouples were spaced apart by stainless-steel washers 5/16 in. O.D. and 1/16 in. thick. Thermocouples were spaced as predetermined for the 5-point probe, 1¼ in. apart, starting 1½ in. from the end of the probe, and for the 4-point probe, at 1 in., 2 in., 4 in., and 5 in. from the end of the probe. The entire probe fit into a stainless-steel protecting jacket 5/16 in. O.D. with .020 in. wall and a turned 30° taper. The jacket was used for protecting the probe when not in use as a method for insertion of the probe into the meat.

The conditions for operation of the experimental oven were determined from computation and varied from test to test. The experimental oven air temperature, circulation, and pressure were continuously recorded or indicated, beginning about 2 hr before the start of the tests and continuing until removal of the test product from the oven. All 3 factors were adjusted by trial and error, since they are interdependent. Upon stabilization of the experimental oven conditions for at least 1 hr, the oven was considered ready for use. The oven door remained closed throughout the test.

The following were measured continually during roasting—time (hundredths of min); oven temperature (6 points in experimental oven, 9 points in control oven, ° F); internal product temperature (9 points, ° F); air circulation in experimental oven (scf); and air pressure in experimental oven (psig).

Upon obtaining a measured low-point temperature of 150° F, the product was removed from the oven and allowed to sit at room temperature for 30 min, the product's internal temperature was recorded, and the thermocouple multiple meat probe cut out, after which the roasted product was weighed (oz) and its volume taken (cc). The roasting pan, rack, and drippings were then weighed (oz). The following computations were made—per cent weight yield, per cent volume yield, per cent evaporation loss, per cent drip loss, and roasting time (min/roasted oz).

RESULTS

General. Per cent weight and volume yield are directly correlated. The correlation coefficient was +.832.

Temperature effect. Per cent weight yield. Absolute and paired difference measurements indicated, in general, that per cent weight yield decreased as temperature increased. The correlation coefficient was −.619. Paired difference measurements indicated that within a limited temperature range, 200–250° F, with definite combinations of circulation and pressure (15 scfm or less and 5–7.5 psig), this general trend may be reversed.

Roasting time (min/oz). Absolute and paired difference measurements indicated that roasting time decreased as roasting temperature increased.

Circulation effect. Per cent weight yield. Absolute and paired difference measurements indicated, in general, that per cent weight yield decreased as air circulation increased. Absolute and paired difference measurements indicated that within a limited air circulation range, 7.5–10 scfm, with a definite combination of temperature and pressure (225° F and 5 psig), per cent weight yield might not decrease, or might decrease and still be significantly high as compared to control conditions (300° F).

Roasting time (min/oz). Absolute and paired difference measurements indicated that roasting time decreased as air circulation increased. This decrease was greatly reduced with an air circulation of over 20 scfm.

Pressure effect. Per cent weight yield. Absolute and paired difference measurements indicated that, depending upon the combinations of temperature, circulation, and pressures used and their levels, pressure may increase, decrease, or have little effect on per cent weight yield. Paired difference measurements indicated that at a pressure of 5–10 psig, with a definite combination of temperature and circulation (225° F and 7.5–10 scfm), significantly high yields may be obtained.

Roasting time (min/oz). Absolute and paired difference measurements indicated, in general, that roasting time decreased as pressure increased up to 10 psig, and that pressures over 10 psig may have little effect on roasting time. Paired difference measurements indicated that at a pressure of 5 psig, with definite combinations of temperature and circulation (225° F and 7.5–10 scfm), roasting time may be increased but still be less than that established for the control oven.

Per cent weight yield vs. roasting time. By using a combination of temperature, circulation, and pressure in the range 212.5–237.5° F, 10–20 scfm, and closed pressure-tight oven to 10 psig (no bleeding), per cent yield may be increased and roasting time decreased.

DISCUSSION

This research indicates that per cent weight and volume yield are correlated. Therefore, in this discussion, when per cent yield is discussed, per cent volume yield is also to be inferred.

Per cent weight yield depends on per cent evaporation loss and per cent drip loss. In this research, per cent weight yield is shown to be inversely correlated with per cent evaporation loss. The correlation between per cent weight yield and per cent drip loss, and between per cent drip loss and per cent evaporation loss, is very poor. McCance and Ship (1933) and many others reported that per cent weight yield is primarily a function of evaporation loss and that water is expressed from meat by contraction of its protein. This research confirms both of these earlier findings. When meat is roasted at low temperatures in a pressure-closed oven (175–225° F), it is common to find water in the drippings upon removal of the meat from the oven. This indicates that water is expressed from the meat although not evaporated. As was expected, the higher the oven temperature, the higher the evaporation loss and the lower the per cent weight yield. Although this is reported (Anon., 1950; Givens, 1955; Lowe, 1946), recommendations have been made within the last year to roast at temperatures of 350° F and above. The results of this research are in agreement with much of the past research and

Table 1. Combinations of temperatures, circulation, and pressures giving best results [a]

Oven air				Change in product yield and roasting time as compared to paired control	
Test no.	Temp (° F)	Velocity (scfm)	Pressure (psig)	Yield (% weight)	Roasting time (min/oz)
20	200	20	10	+8.4	−.29
32	225	15	7.5	+4.4	−.24
33	225	10	5	+5.7	−.22
34	212.5	15	5	+4.2	−.23
38	237.5	10	7.5	+5.6	−.18
37	237.5	15	5	+3.8	−.31

[a] Rounds roasted in control oven at 300° F, natural convection, and atmospheric pressure had an average per cent weight yield of 72.5 ± 6.4 and an average roasting time (min/oz) of 1.7 ± .27 (.05 limits). Combined best results are significantly different from control at .005 level and represent approximately an 8% decrease in roasting losses and a 15% decrease in roasting time.

what has been found to be true in practical cookery; that is, the lower the roasting temperature the higher the per cent weight yield (Anon., 1950; Peyton, 1957). Nothing in this work indicated higher temperatures should be used. To the contrary, it was found that an oven temperature of 250° F gave better yields than the 300° F temperature recommended by most cook-books.

The above indicates that to increase per cent weight yield by decreasing evaporation loss, oven temperature must be lowered. A review of the heat-transfer equations indicates that when oven air temperature is decreased, heat transfer by conduction and radiation will also be decreased. It follows that roasting time will therefore be increased. Heat transfer at low oven temperatures can be increased by forced air circulation. However, this research work also indicates that forced air circulation increases evaporation loss and therefore decreases yield. Clearer understanding of the problem will require study of the factors governing the amount and rate of evaporation of water from meat.

The oven air will tend to reach a state of equilibrium as soon as possible. This condition of equilibrium is that of saturated steam at the pressure of the oven operating temperature. The density of saturated steam or the amount of water needed to produce saturated steam depends on the oven temperature. The total amount of water (moisture) necessary to have saturated steam will depend on the oven temperature and the oven volume. It would therefore seem desirable to have an oven as near the size of the roast as possible. This is true providing the oven is pressure-tight. If the oven is open to the atmosphere, the oven size is immaterial since saturation conditions cannot exist unless the necessary pressure is obtained.

In the case of the experimental TCP oven, the meat had to lose about 10 oz during roasting to saturate the oven air at 300° F. Roasts of the size used met this requirement easily. Smaller roasts would also tend to lose 10 oz or the amount of moisture necessary to saturate the oven air at the oven operating temperature. Of course, this is only true if the oven can obtain the pressure necessary. At lower temperatures, less moisture in necessary to obtain the equilibrium condition; therefore higher yields may result.

Air circulation can be used to make up for the decrease in oven air temperature. However, it has definite limitations. If the air circulation is too high, the roast acquires a "weathered" appearance.

Saturated steam also has the disadvantage of giving the roasted product a moist-heat aroma and texture. It was found that the moist-heat aroma left the product quickly; the texture, however, when operating at high steam pressures, was definitely that of a moist-heat roast.

The obtaining of high yields and low cooking times therefore becomes a balancing task of decreasing evaporation loss by decreasing oven air temperature; increasing heat transfer with forced air circulation to make up for the decrease of oven air temperature, but not enough to give the product the "weathered" appearance; and operating a closed pressure oven to decrease evaporation loss, increase heat transfer by convection, but not at enough pressure to give the product the moist-heat appearance and aroma. The problem is not nearly as simple as some have recently reported, and air circulation alone will not solve the problem and give high yields and low roasting times.

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The Use of Starter Distillate for Flavoring Butter[a]

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(Manuscript received July, 1960)

SUMMARY

The feasibility of flavoring Canadian butter with starter distillate and with a commercial preparation of synthetic diacetyl was investigated at various concentrations. Although official graders preferred control samples to flavored samples, one-third of the members of a taste panel had a definite preference for the flavored samples and only one-fifth preferred the control samples. The remaining panel members showed no preference. Starter distillate butters were preferred to "commercial diacetyl" butters, and 0.50 oz of distillate per 100 lb of fat was preferred to 0.25 oz or 1 oz, whereas for "commercial diacetyl" the preference decreased with increasing concentration.

The levels of acetylmethylcarbinol and of diacetyl in the flavored butters were respectively in the range 0.11–0.27 ppm and 0.24–0.64 ppm. These levels did not change even after 1 year storage at −10° F. Peroxide values indicated that the keeping quality of the butter was unaffected by either flavoring.

IT IS WELL ESTABLISHED that butter made from acid cream has poor keeping quality. The requirements for long storage of butter have led to changes in buttermaking procedures to enhance keeping quality. Some of these changes, such as the use of unripened cream, the standardization of the cream to a low acidity, and possibly the washing of the butter granules, have harmed flavor. Since butter is consumed mainly for its flavor, the question arises whether the bland-flavored butter that results from overemphasis on keeping quality would be preferred by most consumers free to choose a more highly flavored butter.

It is desirable to search for means of enhancing butter flavor without harming keeping quality. Butter flavor can be enhanced by ripening the cream, avoiding excessive neutralization of the cream acidity, eliminating the washing of butter granules, adding starter to cream or butter, or adding flavoring materials such as diacetyl or starter distillate. For keeping purposes, ripened cream butter is out of the question. The standardization of the acidity of butter serum to a pH above 7.0 has never been shown to improve keeping quality. Butter washing has been found (White *et al.* 1956) to be of no help to keeping quality under present Canadian conditions. The presence of starter organisms, even when the factor of acidity is eliminated, has been shown by Wiley *et al.* (1939) to harm the keeping quality of butter. However, recent reports by McDowall *et al.* (1960 a, b) indicated that the use of sweet-cream starter butter does not harm the keeping quality of butter. The use of pure diacetyl has been found to give an unnatural aroma to butter (Babel and Hammer, 1944; Hunziker, 1940; McDowall, 1953; Mohr and Eichstadt, 1935).

Ruehe and Ramsey (1935) improved the aroma of butter by adding starter distillate to either cream or butter. Beck *et al.* (1938) found that this procedure does not affect the keeping quality of butter. Various workers (Babel and Hammer, 1944; McDowall *et al.*, 1960 a, b) have reported that the diacetyl content of butter decreased on storage. No pro-oxidative effect of diacetyl in butter has ever been reported at concentrations below 2 ppm.

Since the manufacture of starter butter requires specialized knowledge, skill, and equipment, it was thought preferable to study more practical means of enhancing butter flavor. The objectives of this study were to determine the stability of some flavor additives in stored butter, their effect on the keeping quality of butter, and whether butter flavored with starter distillate or a commercial diacetyl flavoring would be preferred to conventional butter by a small panel of tasters.

EXPERIMENTAL

Manufacturing. Twenty-seven lots of butter were made from Special Grade cream obtained from the Institute Dairy. They involved three flavoring treatments, three levels of concentration, and three replications. The flavoring treatments were: control, starter distillate, and commercial diacetyl flavoring, and the levels of concentration were 0.25, 0.50, and 1.00 oz per 100 lb of butter. The fat content of the cream was 36%; the acidity, initially 0.11%, was standardized to 0.10% with sodium sesquicarbonate. The cream was pasteurized 10 min at 180° F, cooled to 46° F, and held there until churning. Average churning temperature was 48° F. Just before working, each churning of butter was split into three lots for the following treatments: (a) control, (b) starter distillate added, (c) diacetyl flavoring added. The nine churnings were made at intervals of about three weeks, from May to November, to allow time for the analyses.

The starter distillate and the diacetyl flavoring were both from commercial sources, and both contained 0.11% diacetyl and no measurable quantity of acetylmethylcarbinol. The diacetyl flavoring did not contain any citric, acetic, or propionic acid. Since the loss of diacetyl in the buttermilk is very high (Mohr and Wellm, 1937), the flavoring was added to the butter at the salting stage. The salt content was standardized at 1.9%, and the moisture content at 16%.

Samples from each split churning were saved for the initial analyses. Some 20 one-pound prints from each split churning were wrapped in parchment paper, boxed, and stored at −10° F until examination.

Methods of analysis. Samples from each split churning were scored for flavor initially, at 6 months, and at 12 months, by three government graders who had no knowledge of the experiment. A study of flavor preference was made with an experienced taste panel of 15 people. This panel is regularly used for the appraisal of flavor, odor, or other organoleptic properties of various dairy products. In addition, a detailed procedure of tasting was given to each panelist, and several blank tasting sessions were conducted to train the panel. To reduce the work of the taste panel it was assumed that the three control lots in each replicate were alike in flavor, leaving seven samples for examination in each replicate.

A ranking procedure was used to establish flavor preference. A balanced incomplete block design was used whereby the seven treatments in each of the three replicates were presented to the panel in 35 blocks of 3 units. Each block of samples was ranked

[a] Contribution No. 37.

Table 1. Effect of added flavoring on flavor scores of stored butter (3 replications)

Flavoring per 100 lb butter	Storage at −10° F	Control			Starter distillate			Diacetyl flavoring			Totals
		1	2	3	1	2	3	1	2	3	
0.25 oz	Initial	39	39.5	40	40	39	39	39.5	38	38	352
	6 months	39	39	39	39	39	39	39	39	39	351
	1 year	38.5	40	39	39	39.5	39	39	39	38.5	351.5
0.50 oz	Initial	40	40	39	38.5	39	38	37.5	39	38	349
	6 months	39.5	40	39.5	39.5	39.5	39	38.5	39	39	358.5
	1 year	39.5	39	39	39	38.5	39	39	38	38	349
1.00 oz	Initial	39	40	40	37	39	38	38	38	38	347
	6 months	40	40	39	39	39	39	38.5	39	38	351.5
	1 year	39	39	39	39	38.5	39	39	38	38	348.5
	Totals	353.5	356.5	353.5	350	357	349	348	347	344.5	

from 1 to 3 in order of preference for flavor by each member of the taste panel. Each treatment appeared five times in the same block with each other treatment.

Samples from each lot were analyzed for diacetyl and acetylmethylcarbinol at 0 and 3 days; 1, 2, 3, and 5 weeks; 2 months; and 1 year, according to the method of Prill and Hammer (1938). Assuming 100% purity for reagent grade diacetyl, the diacetyl recovery was 93%. For determinination of the combined diacetyl and acetylmethylcarbinol, 20 ml of 40% ferric chloride were added to 100 g of butter before distillation, and the degassing with nitrogen was omitted. Peroxide values were determined by the method of Lips *et al.* (1943).

RESULTS AND DISCUSSION

Flavor score. The flavor scores given by Canadian official graders in Table 1 and analyzed for significance in Table 2 indicate that the graders preferred the control samples to the flavored butters, and the starter distillate butters to the diacetyl flavoring butters. As expected, preference was in the inverse order of concentration. The 6-month samples received a higher average score than the fresh or the 1-year samples. The flavor preference is indicative of the trend in Canada for official graders to favor butter with "bland" flavor. McDowall (1953) reported a similar observation with New Zealand butter containing 0.05 ppm diacetyl. The New Zealand graders criticized the butter as harsh and "overfull" in flavor.

Table 2. Analysis of variance of flavor scores

Source of variation	Degrees of freedom	Mean square
Flavoring	2	5.36[a]
Control vs flavoring	1	8.68[a]
Starter dist. vs commercial diacetyl	1	2.05[b]
Concentration	2	0.53
Linear	1	1.04[b]
Quadratic	1	0.02
Storage	2	0.70[b]
Linear	1	0.02
Quadratic	1	1.39[b]
Flavoring × concentration	4	0.47
Flavoring × storage	4	0.58[b]
Concentration × storage	4	0.53
Flavoring × concentration × storage	8	0.15
Replication	2	0.53
Error	52	0.22

[a] Significant at P = 0.01.
[b] Significant at P = 0.05.

Taste panel. Table 3 shows the rank totals for the 15 panelists. The over-all analysis of variance of the normal varieties (Fisher and Yates, 1953) with the treatment totals adjusted according to Cochran and Cox (1957) indicates that the difference between treatments is almost significant at P = 0.05. The adjusted treatment mean square on a unit basis was 2.04, against a P = 0.05 of 2.24. The concordance coefficient (Kendall, 1948) indicated also that the members agreed on a difference between treatments.

Since taste preference is known to vary with individuals, an over-all analysis of variance of preference data could be misleading when there are reverse preferences by substantial portions of the panel. To obtain the full information from the preference data, the members were grouped on the basis of the significance of the chi-square values of their preference. Table 4 indicates that five members preferred the

Table 3. Flavor ranking of butters flavored with starter distillate or diacetyl flavoring. (Each value[a] is the sum of 15 judgments by taste panel members)

Block no.	Control	Starter distillate (oz) per 100 lb of fat			Diacetyl flavoring (oz) per 100 lb of fat		
		0.25	0.5	1	0.25	0.5	1
1	27			28			35
2	30		29	31			
3	29			28		33	
4	32	29		29			
5	35			25	30		
6	35		26				29
7	35					25	30
8	28	28					34
9	32				26		32
10	32		26			32	
11	31	27	32				
12	32		31		27		
13	30	26				34	
14	30				32	28	
15	31	29			30		
16			27	30			33
17				34		26	30
18		25		30			35
19				27	30		33
20			29	32		29	
21		26	29	35			
22			29	29	32		
23		30		27		33	
24				30	30	30	
25		28		29	33		
26			27			32	31
27		30	27				33
28			26		28		36
29		29				29	32
30					34	27	29
31		30			27		33
32		30	26			34	
33			28		32	30	
34		27	32		31		
35		31			27	32	
Totals	469	425	424	444	449	454	485
Adjusted totals	457.2	440.6	440.2	447.7	449.6	451.5	463.2

[a] Increasing values = decreasing preference.

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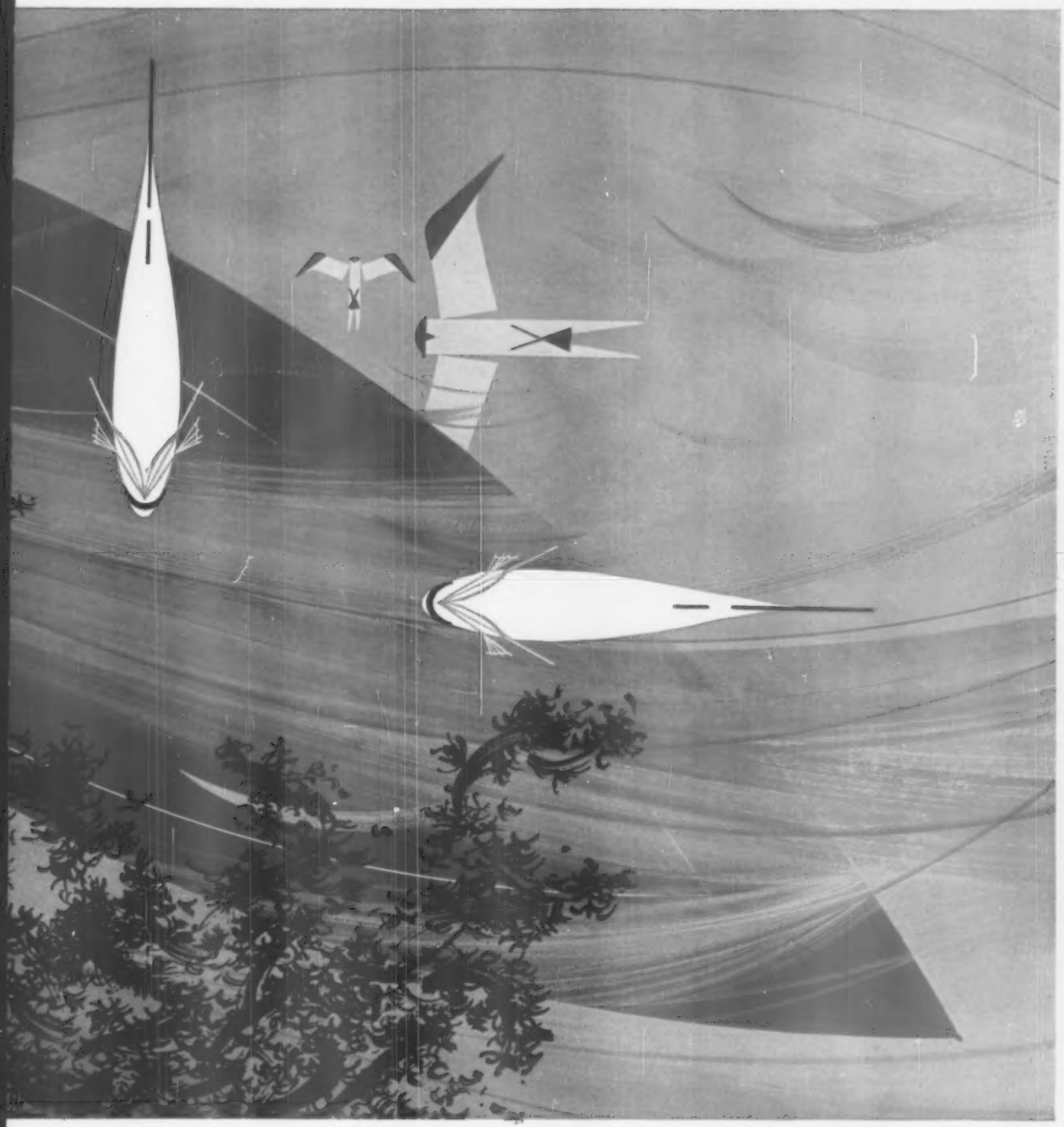

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Table 4. Grouping of panel members according to their flavor preference

Panelists	Treatment						Chi-square
	B	C	D	E	F	G	
Preferring flavored butters	No. times in five the treatment was preferred to the control.						
1	5	4	4	4	2	1	12.4
2	4	5	4	4	5	5	20.4
3	4	3	2	4	5	5	14.0
4	3	5	5	4	3	3	12.4
5	5	5	5	5	4	5	26.8
Preferring control sample							
6	0	1	2	0	2	2	12.4
7	0	0	3	1	3	2	12.4
8	0	1	1	0	1	0	20.4
Showing no preference							
9	3	3	2	4	1	2	4.4
10	2	3	1	1	1	3	6.0
11	4	1	4	3	3	4	7.6
12	3	3	0	4	3	3	7.6
13	3	1	3	0	3	2	7.6
14	3	2	2	2	4	1	4.4
15	1	2	3	4	3	2	4.4

flavored samples to the control, three preferred the control to the flavored samples, and seven showed no preference.

For the panelists showing an over-all preference for the flavored butters, the analysis of variance of the normal variates after adjustments for the treatments for incomplete block analysis (Cochrane and Cox, 1957) showed a highly significant difference between treatments. The multiple range test of Duncan (1955) indicated that the members preferred the flavored butters over the control but had no preference among the flavored butters, varying in flavoring type and in flavoring concentration.

For the three members showing an over-all preference for the control over the flavored samples, the analysis of variance showed a significant difference between the adjusted treatments. The multiple range test of Duncan established that these panelists did not distinguish between the control and the low level of starter distillate, but preferred these to all other treatments. Among the flavored samples, the low and medium levels of starter distillate were preferred to the samples with diacetyl flavoring or with high level of starter distillate.

Acetylmethylcarbinol. The average acetylmethylcarbinol content of the control samples was 0.11 ppm. This value is an agreement with the value of 0.16 ppm reported by Mohr and Wellm (1937) for sweet-cream butter. The acetylmethylcarbinol content of the flavorings used was not measurable by the method used. However, the addition of flavorings to butter effected a slight increase in the acetylmethylcarbinol content, more so for commercial diacetyl than for starter distillate. Even at the rate of 1 oz of flavoring per 100 lb of butter the acetylmethylcarbinol content was raised only to 0.17 ppm and 0.21 ppm for starter distillate and for commercial diacetyl, respectively. The variates storage and concentration had no effect on the acetylmethylcarbinol content of the butter.

According to Hedrick and Hammer (1942), starter butter may contain as much as 16 ppm of acetylmethylcarbinol. McDowell *et al.* (1960 a, b) obtained 1.4–3.9 ppm in their sweet-cream starter butter. They found that storage for 8 months at 14° F effected a slight increase (8–15%), significant at P = 0.01, in the acetylmethylcarbinol content of the butter. The increase was not significant after 4 months. Presence of starter and relatively high storage temperature may have accounted for this slight increase.

Diacetyl. The diacetyl content of the control samples averaged 0.16 ppm. This value is in the range of those reported by other workers for sweet-cream butter. Cox (1942) found 0.02–0.20, average 0.07 ppm, Barnicoat (1937) 0.05 ppm, and Mohr and Wellm (1937) 0.2 ppm.

Starter butters can have as much as 2 ppm diacetyl, as in Danish butter (Pette, 1948), or have less than 0.5 ppm, as in most French butters (Pien *et al.*, 1937). According to Barnicoat (1937) and Davies (1937), the diacetyl content of mild starter butter is about 0.4 ppm, and that of "high-acid" butter is 1.5 ppm. Davies classified butters on the basis of their diacetyl content as follows: 0.2–0.6 ppm is mild, 0.7–1.5 ppm is full, and 2 ppm is strong. With this classification, the flavored butters in our experiment would fall into the category of mild (Tables 5 and 6).

On adding flavoring to butter, there was a loss of about 50% of diacetyl. This percentage loss increased with the level of added flavoring. McDowall (1953) found a similar loss, which he attributed to loss to the atmosphere and to wash water during churning. Since the flavoring was added to the butter at the

Table 5. Diacetyl content (ppm[a]) of stored butter flavored with starter distillate

Storage at −10° F	Control [b]	Oz of added distillate per 100 lb of butter			Mean
		0.25	0.50	1.00	
Fresh	0.14	0.26	0.30	0.42	0.28
3 days	0.15	0.25	0.30	0.43	0.28
1 week	0.16	0.26	0.29	0.44	0.29
2 weeks	0.14	0.20	0.28	0.38	0.25
3 weeks	0.17	0.24	0.31	0.40	0.28
5 weeks	0.18	0.26	0.32	0.42	0.29
8 weeks	0.18	0.26	0.31	0.40	0.29
1 year	0.15	0.24	0.29	0.43	0.28
Mean	0.16	0.25	0.30	0.41	0.28

[a] Individual values are averages of three replicates.
[b] Control values are averages of nine churnings.

Table 6. Diacetyl content (ppm[a]) of stored butter flavored with diacetyl flavoring

Storage at −10° F	Control [b]	Oz of added distillate per 100 lb of butter			Mean
		0.25	0.50	1.00	
Fresh	0.14	0.32	0.44	0.61	0.38
3 days	0.15	0.32	0.45	0.59	0.38
1 week	0.16	0.28	0.42	0.64	0.38
2 weeks	0.14	0.27	0.42	0.56	0.35
3 weeks	0.17	0.28	0.45	0.60	0.38
5 weeks	0.18	0.29	0.43	0.58	0.37
8 weeks	0.18	0.30	0.38	0.57	0.36
1 year	0.15	0.26	0.42	0.55	0.34
Mean	0.16	0.29	0.43	0.60	0.37

[a] Individual values are averages of three replicates.
[b] Control values are averages of nine churnings.

salting stage in our experiment, wash water was not a factor. However, the diacetyl could have disappeared by volatilization and by oxidation or reduction.

Tables 5 and 6 show the diacetyl content of butter for both flavorings at various levels. Table 7 indicates

Table 7. Analysis of variance of diacetyl values

Source of variation	Degrees of freedom	Mean square
Flavoring	1	0.437[a]
Concentration	2	1.161[a]
Storage	6	0.005
Flavoring × concentration	2	0.126[a]
Flavoring × storage	12	0.085
Flavoring × concentration × storage	12	0.098[a]
Error	84	0.075

[a] Significant at P = 0.01, using the appropriate significant interaction to test for flavoring, concentration, and storage.

that in spite of the equivalent strength of the flavorings, more diacetyl was imparted to butter by commercial diacetyl than by starter distillate. This is substantiated by the significant interaction between flavoring and concentration.

Table 7 also shows that storage for 2 months or 1 year at −10° F did not change the diacetyl content of the butter. In two published papers, McDowall (1960 a, b) found that storage of sweet-cream starter butter at 14° F decreased the diacetyl content by 14–20% in 4 months. Analysis shows their data were significant at P = 0.01. Prolonged storage for 8 months did not change the diacetyl content. The authors did not mention whether the butter was salted. The presence of microorganisms, along with the relatively high storage temperature and possibly a low salt content, probably accounts for the decrease in diacetyl content during storage. As reported by Babel and Hammer (1944), many other workers have observed that the diacetyl content of butter decreased on storage; this was generally attributed to the reducing action of microorganisms or to oxidation by air, especially in the presence of acid or in the absence of added salt. With salted butter, several authors have reported that the diacetyl was more stable.

The keeping quality of the butter was not affected by the presence of flavorings, as indicated by the absence of any difference in peroxide values between the treated and the control samples after 1 year of storage at −10° F.

Acknowledgment

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Wheat Gliadin in Foams for Food Products[a]

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SUMMARY

Aqueous solutions of gliadin, partially deamidized gliadin, and gliadin partially hydrolyzed by pepsin can be whipped into stable foams similar in some respects to those produced from egg white. At slightly acid pH's, foams from gliadin and partially hydrolyzed gliadin were similar in volume, but those from the former had somewhat greater stability. At pH 5 the volume and stability of deamidized gliadin foams were equal to those of unmodified gliadin, but at higher pH's, foam volumes were smaller and less stable. Deamidized gliadin is compatible with egg white. Mixtures of the two gave foams of greater volume and stability than those from mixtures of egg white and gliadin or partially hydrolyzed gliadin. Pie meringues containing gliadin or deamidized gliadin were excessively soft and unstable at oven temperatures. Deamidized gliadin alone, or as a mixture with egg white, made excellent meringue shells, divinity candy, fluffy cake icings, and stabilized butter cream cake icings, whereas unmodified gliadin performed poorly in these food products. These findings establish the essential suitability of foams from gliadin or its derivatives for use in a variety of food products.

AQUEOUS SOLUTIONS of gliadin, the alcohol-soluble fraction of wheat gluten, can be whipped into stable foams similar in many respects to those from egg white. The relative abundance and low cost of wheat gluten focuses attention on the possible use of gliadin in food foams (Sagi, 1954). Large quantities of food foams are used each year in pie meringues, angel food cakes, dessert toppings, candies, and the like. Most such products are made fairly satisfactorily with egg white, gelatin, soy proteins, or modified milk proteins. Thus, any new material would have to exhibit superior performance at no increase in cost, or equal performance at less cost.

This study is concerned with the foaming properties of high-quality gliadin or gliadin derivatives, but not with relative costs. Egg white was used as a reference material because of widespread familiarity with its foaming properties.

Gliadin, about one-half the protein material in crude gum gluten, is extractable from it with aqueous alcohol solutions. Rather recently it was shown to consist of a mixture of at least four electrophoretically distinct protein components (Jones *et al.*, 1959), but the traditional term for the mixture is retained for convenience.

MATERIALS AND METHODS

Preparation of Gliadin. Gliadin was extracted from commercial dried gum gluten (75% protein) with aqueous 35% (v/v) isopropanol at room temperature using 9 ml of alcohol solution per gram of gluten. After 45 minutes of stirring, the insoluble material was collected by centrifugation to leave a cloudy supernatant having a pH of about 5.8. In about three hours at 22°C, after the pH had been adjusted to about 7.0, the finely dispersed material precipitated well enough to be removed by centrifugation. Lipid impurities were next removed by stirring the supernatant for an hour with 16 g/L of Norite A[c] at a pH adjusted near 5.9. A water-clear solution was then obtained, after addition of 16 g/L of filter aid, by filtering on a filter-aid mat on a Buchner funnel. The gliadin was precipitated from the alcoholic solution by adjusting it again to pH 7.0, warming to 40°C, diluting with one-half volume of water, and cooling to 10°C. The precipitate was recovered either by centrifugation or by decantation after standing overnight at 5°C. Washing the precipitated material several times at 50°C with distilled water removed traces of impurities and isopropanol. After dispersing the precipitated gliadin in 0.1*N* acetic acid, it was recovered by freeze drying. On a dry-weight basis the product contained 17.7% nitrogen, of which 25.6% was amide nitrogen.

Preparation of Deamidized Gliadin. Deamidized gliadin was prepared by dispersing the final gliadin precipitate in 0.07*N* hydrochloric acid at a concentration of about 5.5% and heating 3 hr at 96–98°C. This treatment removed about 28% of the total amide groups as indicated by an increase in ammonia determined by direct analysis of the hydrolysate with a ninhydrin reagent (Moore and Stein, 1948). Ammonia is believed liberated under the hydrolysis conditions without significant splitting of peptide bonds (Holme and Briggs, 1959, Vickery, 1922). The deamidized material was recovered by adjustment of the pH to 3.9. After decantation the precipitate was suspended in water, and its pH was adjusted to 5.3 before it was dried by lyophilization. The final product contained 16.4% nitrogen, of which 21.7% was amide nitrogen.

Partially Hydrolyzed Gliadin. About 52 mg of pepsin (Armour and Company, 1–10,000) were added to each liter of gliadin solution at pH 2.5 and approximately 11% concentration of protein. The mixture was allowed to react at 28°C for 20–25 minutes, during which time about 0.15% of the total peptide bonds were hydrolyzed, as determined by titration (Jacobsen *et al.*, 1957). Action of the enzyme was stopped by raising the pH to 4.7 and heating the solution to 90°C for one minute. The protein material was then recovered by freeze-drying.

Frozen Egg White. A uniform supply of frozen egg white was prepared by blending fresh whites and freezing in small aliquots to be held until needed. Prior to freezing the whites were stirred in an electrical blender at low speed until the drain time from a No. 2 Zahn viscosimeter was less than 20 seconds. Incorporation of air during blending was minimized by partial immersion of an upright Erlenmeyer flask into the mixing vortex.

Foam Preparation. Foams were prepared with a Hobart K-4-B mixer operated at maximum speed and fitted with a wire batter-whip. Speed uniformity was checked daily with a clutch-type speed counter (Veeder-Root, Inc., Hartford) after a warm-up of 15–20 minutes. Test samples consisted of 60 ml of protein solution in which the various additives were suspended or dissolved.

Foam expansion values were determined as the ratio of final foam volume to original liquid volume by calculation from the weight of foam required to fill a quarter-cup measure (59 ml).

Foam stability at room temperature was measured in terms of the leakage of liquid from a fixed volume of foam over a two-hour period. A half-cup measure (118 ml) filled with foam

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[b] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, United States Department of Agriculture.

[c] Mention of firm names or trade products does not constitute endorsement by the U. S. Department of Agriculture over others of a similar nature not mentioned.

was covered with a tinned wire screen held in place by a narrow glass plate clamped across the midsection of the screen. The assembly was mounted on a ring stand with a burette clamp, so that free liquid drained into a 10-ml graduated cylinder fitted with a funnel. Stability was expressed as the per cent, by weight, of liquid draining from the foams.

Both measurements were run in duplicate and were conducted in an air-conditioned room maintained at 23 ± 0.5°C, unless otherwise noted.

RESULTS AND DISCUSSION

Preliminary testing indicated that non-nitrogenous impurities in gliadin preparations prolonged the mixing required to attain maximum foam volume, and reduced the foam stability. All experiments reported were therefore conducted with the high-purity preparations just described.

Effect of Beating Time. As indicated in Fig. 1, foams formed quickly upon beating 3% solutions of unmodified gliadin. Foam expansion reached a maximum in about 4 minutes, whereas foam stability increased for the first 6 minutes and decreased only slightly on further beating.

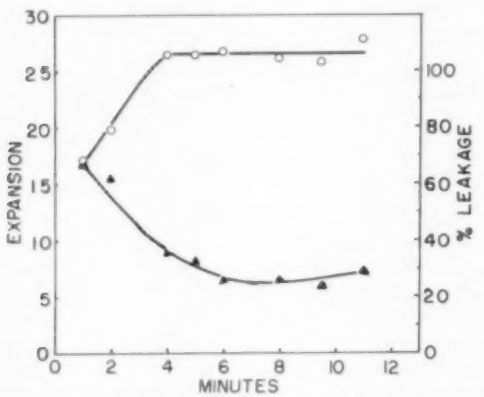

Fig. 1. Expansions and leakages of foams produced from 3.0% solutions of unmodified gliadin (pH 4.2). Open circles, foam expansions; solid triangles, foam leakages.

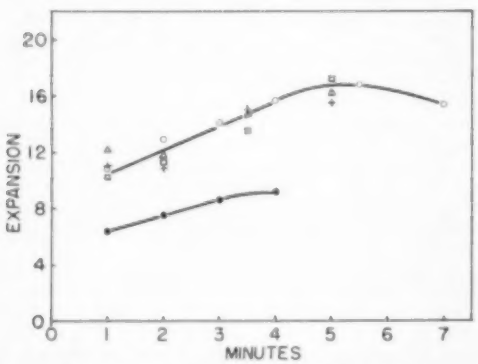

Fig. 2. Expansions of foams produced from 3.1% solutions of unmodified gliadin and from egg white containing 1.3% cream of tartar, each containing 35 g sucrose per 60 ml of protein solution. Gliadin solutions pH 3.3, open squares; pH 4.0, open triangles; pH 4.7, open circles; pH 5.2, cross. Egg white, solid circles.

Effect of pH. Sucrose-containing gliadin solutions (3%) at pH values ranging from 3.3 to 5.2 gave similar foam volumes, as shown by the upper line in Fig. 2. Egg white (natural pH 9.0) containing cream of tartar added just prior to beating (final pH near 5) gave a similar curve but at a lower expansion range. The lower expansions of egg are presumably due to its high solids content (12%) (see also Table 1).

As indicated in Fig 3, solutions of deamidized gliadin (3%) ranging in pH from 5.2 to 7.5 and containing sucrose produced smaller foam volumes at the successively higher pH values. At pH 5.2, foam expansions were equal to those observed with unmodified gliadin (Fig. 2).

Foam volumes for the solutions of partially hydrolyzed gliadin (3%) containing sucrose differed little at various pH values, as shown in Fig. 4. These re-

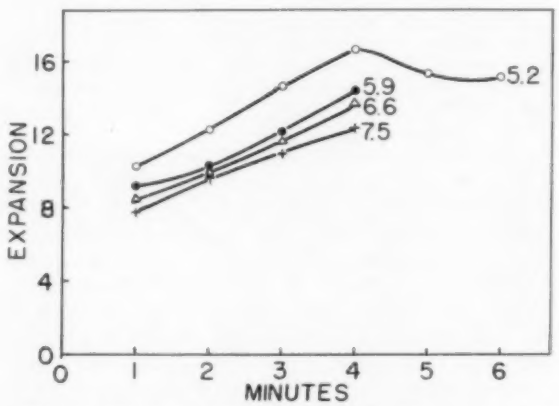

Fig. 3. Expansion ratios for foams produced from 3.1% solutions of deamidized gliadin (35 g sucrose per 60 ml protein solution). The pH of the gliadin solution is indicated by the number next to each curve.

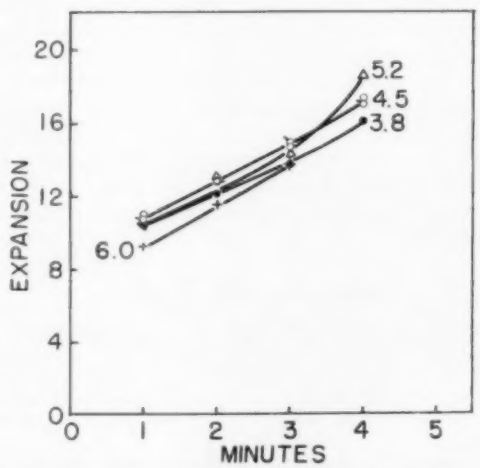

Fig. 4. Expansion ratios for foams produced from 3.1% solutions of partially pepsin-hydrolyzed gliadin (35 g sucrose per 60 ml protein solution). pH values are indicated by the curves.

sults are similar to those obtained with unmodified gliadin solutions (Fig. 2).

Foam stabilities at several pH values for gliadin solutions (3%) containing sucrose (Fig. 5) reached a maximum after 2–4 minutes of beating, and remained constant on further beating. In contrast, egg white (undiluted) containing cream of tartar and sucrose reached maximum stability in one minute or less, and decreased on continued beating.

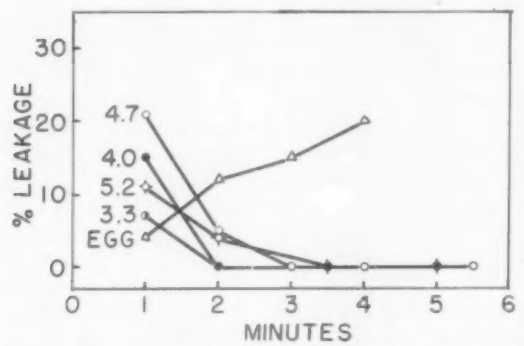

Fig. 5. Leakages after two hours from foams produced with 3.1% solutions of unmodified gliadin and from egg white containing 1.3% cream of tartar each containing 35 g sucrose per 60 ml protein solution. The numbers next to each curve give the pH of the gliadin solutions.

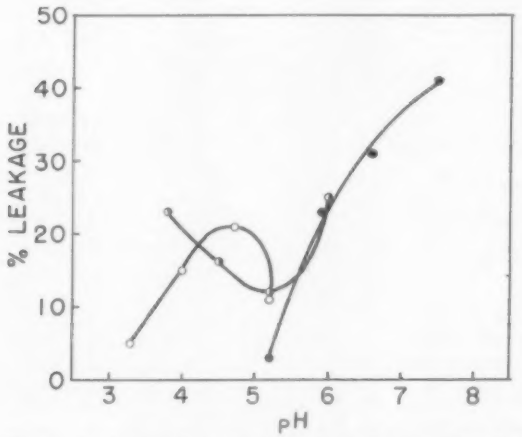

Fig. 6. Leakages after two hours from foams produced by 1 minute of beating 3.1% solutions of gliadins at different pH's and containing 35 g sucrose per 60 ml of protein solution. Open circles, unmodified gliadin. Half solid circles, pepsin-treated gliadin. Solid circles, deamidized gliadin.

Fig. 6 gives the leakages for foams from the different gliadin-sucrose solutions after one minute of beating. Foams from all of them appeared to have a maximum stability near pH 5.2. Those from unmodified gliadin approached another maximum near pH 3, whereas those from partially hydrolyzed gliadin decreased in stability at pH's above and below 5.2. Deamidized gliadin foams decreased in stability with increasing pH.

Effect of Additives. Tables 1–3 show the effect of protein concentration and several additives on foam volumes and stabilities for solutions of unmodified gliadin beaten for 7 min.

Table 1. Effect of protein concentration on properties of foams from unmodified gliadin at 25° C.

Gliadin concentration	Expansion ratio	Leakage
%		%
1.5	22.0	55
3.0	25.8	32
4.6	23.0	14
6.1	17.5	5
9.2	10.6	0

Table 2. Effect of sucrose and salt on foam properties of 3.0% solutions of unmodified gliadin at 25° C.

Additive	Additive concentration	Expansion ratio	Leakage
	%		%
Sucrose	0	25.8	32
	14.3	24.5	18
	25.0	23.2	9
	33.3	20.2	2
	40.0	14.7	0
Sodium chloride	0.06	25.5	14
	0.11	24.1	14
	0.17	16.4	37

Table 3. Effect of lipids on foam properties of 3.0% solutions of unmodified gliadin at 25° C.

Additive	Additive concentration	Expansion ratio	Leakage
	%		%
Cottonseed oil	0	25.8	32
	0.94	21.8	59
	2.20	17.1	71
	3.00	14.5	81
Soy phosphatides, alcohol-soluble	0.09	25.3	66
Soy phosphatides, alcohol-insoluble	0.09	26.0	58

Table 1 shows that 3% gliadin solutions gave maximum foam expansion, but foam stability varied directly with protein concentration. Table 2 shows that sucrose enhanced stability but decreased foam volume. Salt interfered by insolubilizing the gliadin (Table 2). A 0.11% concentration of salt caused the appearance of particles of precipitated gliadin, and 0.17% prevented complete dispersion of the gliadin prior to beating. Cottonseed oil (Table 3) caused reduction in both foam volume and stability, whereas soy phosphatides at the concentration used reduced only the stability.

Table 4 shows expansions and leakages of foams formed at pH 5.0 by beating for 1 and 2 min. gliadin and modified gliadin solutions (3%) containing 50% added egg white (undiluted) as well as sucrose. Gliadin itself precipitated on mixing with egg white. This precipitate greatly delayed foam formation. The modified gliadins were not subject to this trouble. The deamidized gliadin–egg white mixture gave the greatest volume and the most stable foams. The partially hydrolyzed gliadin–egg white mixture was no better than egg white diluted with water. The relatively rapid breakdown of the diluted egg foams is

Table 4. Foam properties of egg white–gliadin mixtures and of egg white at pH 5.0.[a]

Sample			Foam properties	
Volume of 3.1% solution of gliadins (ml)	Volume of egg white (ml)	Beating time (min.)	Expansion ratio	Leakage (%)
30, unmodified	30	2.0	8.4	12
30, pepsin treated	30	1.0	7.1	36
30, pepsin treated	30	2.0	10.0	8
30, deamidized	30	1.0	9.3	2
30, deamidized	30	2.0	11.4	0
0	30[b]	1.0	7.1	27
0	30[b]	2.0	10.0	19
0	60	1.0	4.7	21

[a] Egg white was adjusted to pH 5.0 with 2.0*M* citric acid. Each sample contained 35 g sucrose.
[b] 30 ml of water also added to obtain the 60-ml test volume used.

presumably due to the unfavorable acid conditions (pH 5) required for solubilization of the gliadins.

Performance in Food Products. Pie meringues were prepared with 10% solutions of deamidized gliadin (pH 5.2), egg white, or 50:50 mixtures of the two. Meringues made with deamidized gliadin, or 50:50 egg white and deamidized gliadin, required a longer time to brown on lemon pie fillings than did all-egg meringues. Their texture in comparison to all-egg meringues was excessively soft and moist, but the ridges and peaks stood up well. This softening effect was noticeable, though not objectionable, with as little as 10% deamidized gliadin added to egg white.

Meringue dessert shells of excellent quality were prepared with foams from 10% solutions of deamidized gliadin at pH 5.2 or 8.3, or from 50:50 mixtures of these solutions with egg white. Shells containing deamidized gliadin required a slightly longer baking time but were much lighter in color. Shells from the gliadin were almost snow white, whereas those from egg white were light tan, probably because of interaction of egg proteins with glucose naturally present in egg white. The low content of amino groups in gliadin also causes it to brown more slowly. Foams from a 10% dispersion of unmodified gliadin at pH 4.7 produced shells that were so fragile they could not be removed from the baking sheets without breakage.

A 10% solution of deamidized gliadin was also used as a stabilizer in butter cream cake icing to prevent graininess due to sugar crystal growth. The icing had a high gloss and was creamy in texture. At pH 8.3 the deamidized gliadin prevented graininess for only 1–2 days, but at pH 5.2 it performed as well as egg white and stabilized the icing for at least 11 days. At both pH's, 50% deamidized gliadin–egg white mixtures prevented graininess over the same period. A 10% solution of unmodified gliadin produced an icing with a dull gloss and rough texture; after four days, water leaked from the icing.

Deamidized gliadin produced a slightly stiffer consistency and greater volume than egg white when used in uncooked fluffy cake icings prepared by adding boiling water to a dry mix and beating (Butler, 1959). The deamidized gliadin icings were equivalent to those from egg white in color, flavor, and stability. An attempt to prepare this icing with unmodified gliadin failed because it was made insoluble by the salt present and could not be whipped.

Divinity candy prepared from deamidized gliadin was slightly smoother than candy made from egg white immediately after preparation. After two weeks in a closed container, both appeared nearly equal in texture. Divinity candy prepared with unmodified gliadin was heavy and fondant-like because of breakdown of the gliadin foam when the hot syrup was added.

In all the food products used, excepting the pie meringues, deamidized gliadin performed as well as egg white, or better. Unmodified gliadin performed poorly under most conditions. The better compatibility with egg white and the better performance of deamidized gliadin, in general, make the latter product of greater interest for commercial adaptation.

The flavor of the gliadins used in these experiments was bland enough that no intrusion or interference with flavors imparted by other ingredients in the food products could be noticed, although panel evaluations were not conducted. Pie meringues made with gliadin or its derivatives seemed somewhat flat in taste when compared with egg meringues. Such an effect was much less noticeable with other products containing higher levels of sugar or other flavorings. Another difference in organoleptic properties was the lighter body of foams made with the gliadins. This effect is probably due to a combination of relative lack of flavor from the protein and a more moist "mouth feel" than that characteristic of egg white foams.

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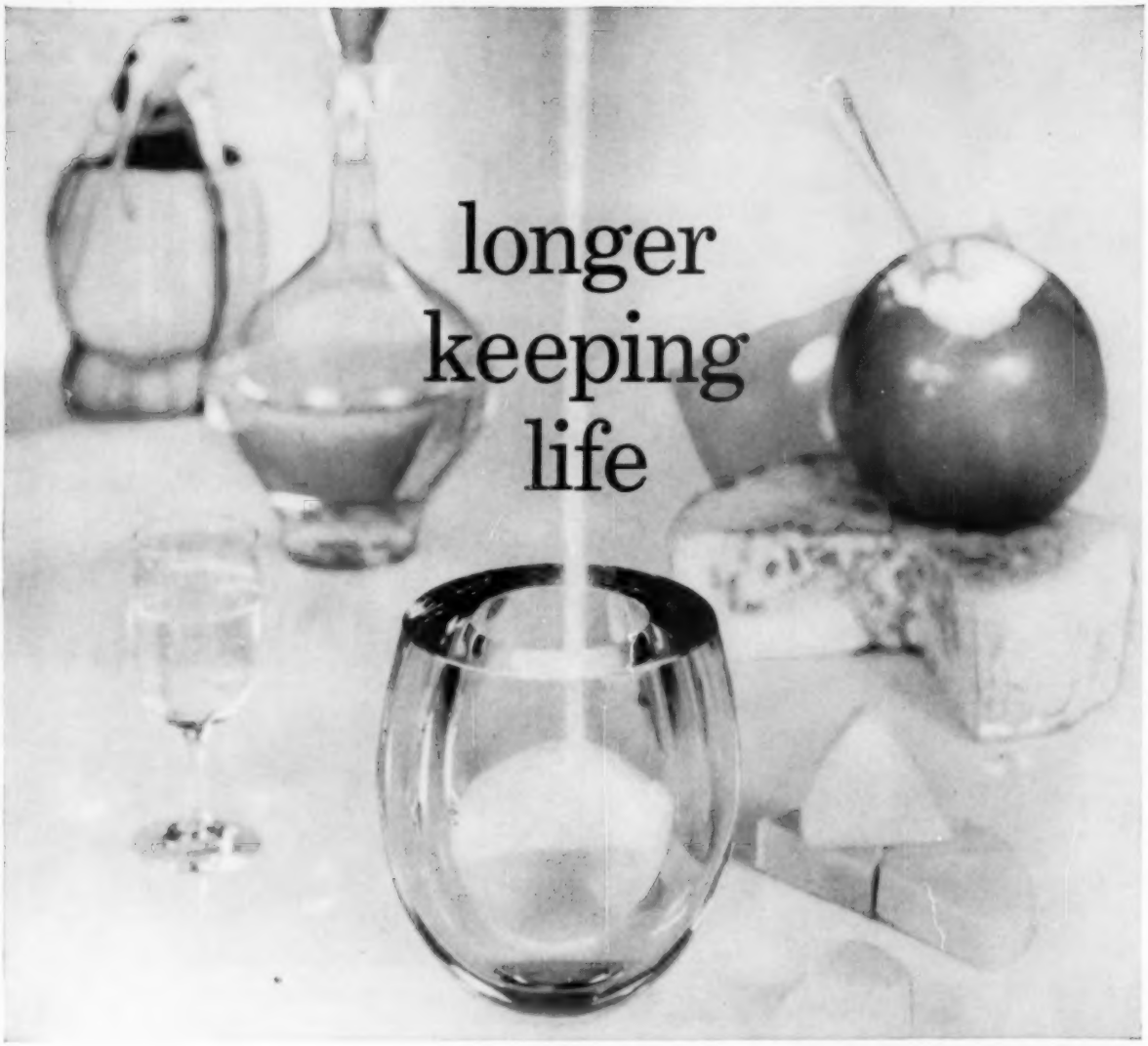

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144B

Preservation of Fish and Shellfish by Relatively Low Doses of Beta Radiation and Antibiotics[a, b]

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(Manuscript received June, 1960)

SUMMARY

The complementary action of a treatment with chlortetracycline and beta irradiation on the retardation of spoilage of flatfish and rockfish fillets, salmon steaks, precooked tuna loins, cooked crabmeat and shrimp has been studied over a range of doses from 11,800 to 280,000 rads. The additional effect of an antioxidant treatment on salmon preservation has been exemplified. Preliminary studies on the effect of beta irradiation on the viability of selected bacterial strains from fresh and spoiling fish have also been carried out. The relative sensitivity of Micrococci, Flavobacteria, Achromobacter and Pseudomonas species to beta radiation has been verified.

A RATHER EXTENSIVE literature has accumulated on the sterilization of foods and related commodities by beta and gamma radiations from various sources. Among the beta radiation sources used have been Van de Graaf electron accelerators, resonant transformer electron-beam generators, and linear electron accelerators. The source of the gamma radiation has usually been radioactive cobalt 60. The majority of the studies have used high doses of radiation and have been concerned with producing complete sterilization, the final products needing no refrigerated storage conditions. Even though sterile products with storage capabilities at ambient temperatures have resulted from high-dose irradiation, the general consensus was that many of them had a limited acceptability because of the development of the so-called "radiation odors and flavors." The basic principles and possible applications of the irradiation of foodstuffs have been described in a number of papers (Anon., 1954; Ede, 1950; Goldblith and Proctor, in 1955; Hannan, 1955; Huber, 1951; Lück and Kuhn, 1959; Symposium, 1954).

With respect to fish preservation the majority of the studies have been concerned with the effects of high doses of millions of rads (Gardner and Watts, 1957; Hannan, 1955; MacLean and Welander, 1960; Miyauchi, 1958; Nickerson, 1954; Nickerson *et al.*, 1950; Okada *et al.*, 1955; Proctor and Bhatia, 1950; Proctor *et al.*, 1950). The general consensus has been that fatty fish, such as mackerel and herring, do not withstand these high doses of irradiation, and among other changes become rancid. The low-fat white-flesh fish, such as cod and haddock, gave products with somewhat better acceptabilities after being treated with 1–3 megarads.

Relatively fewer studies on foods, including fish, have been carried out with low or relatively low doses of irradiation ranging from about 100,000 to less than a million rads. The possibilities of this type of irradiation treatment for foods have been discussed in an editorial (Anon., 1954) and by Eukel and Huber (1960) and Goldblith and Proctor (1955). Gardner and Watts (1957) found that spoilage of oysters was not retarded by doses of gamma radiation from 63,000 to 630,000 rads. They concluded that radiation preservation of oysters in the raw state did not hold great promise even at refrigerator temperature (5°C). Liston and Shewan (1957) found some flavor deterioration in cod fillets after gamma radiation doses of 250,000 to 1 million rads and storage at 0°C. Chemical determinations of volatile bases did not correlate with the sensory judgments. Irradiating cod and lemon sole, while frozen, with 250,000 and 500,000 rads of beta radiation gave fillets that were rated almost as highly as unirradiated controls. In contrast to these results, mackerel and herring were considered unacceptable even after a dose of 250,000 rads of beta radiation while in the frozen state. Shewan and Liston (1959) also concluded that low doses of irradiation were most suitable for such fish as cod and sole. Retardation of spoilage and prolongation of storage life at low temperatures above freezing also have been reported for cod fillets, smoked fish, and shellfish (MacLean and Welander, 1960; Miyauchi, 1958), for haddock (Nickerson *et al.*, 1954) and for tuna and fish pastes (Okada *et al.*, 1955). Some preliminary results have also been reported (Eukel and Huber, 1960) on the effect of low doses of beta radiation on fillets of sole and of rockfish, and on salmon steaks.

The increased efficacy of a combined treatment with relatively low doses of gamma radiation and oxytetracycline, compared to the action of irradiation alone, on the keeping quality of beef was first reported by Niven and Chesbro (1957). Similar beneficial effects of a combined treatment with gamma radiation and chlortetracycline on the spoilage retardation of cod fillets were reported by Liston and Shewan (1960) and by Shewan and Liston (1959). Some public

[a] Presented at the 20th Annual Meeting of the Institute of Food Technologists, San Francisco, California, May 15–19, 1960.

[b] Based on Ph.D. studies carried out by Peter Lerke at the University of California, 1958.

health aspects of the irradiation of food have been discussed by Nickerson *et al.* (1953).

In this report are presented data on the effect of relatively low doses of beta radiation, ranging from 11,800 to 280,000 rads, alone and in combination with 5–25 ppm of chlortetracycline (CTC) on the rates of spoilage of fillets of sole and of rockfish, of salmon steaks, of tuna loins, and of commercially cooked crabmeat and shrimp during storage at household refrigerator temperatures (42°F or 5.5°C) and at 35°F (1.7°C). Also included are some data on the effect of doses of beta radiation up to several million rads on the color of salmon steaks and on the radiation sensitivity of various genera of bacteria isolated from raw fresh and spoiled fish.

MATERIALS AND METHODS

Source of radiation. The radiation source was the linear electron-beam accelerator facility of the Applied Radiation Corporation, Walnut Creek, California. The beta radiation source was at an energy level of 5–6.5 Mev. The doses were standardized by the cobalt blue glass dosimetry procedure.

Fish and shellfish. The fillets of sole (Dover, Petrale, English, all varieties of flatfish of the flounder family) and of rockfish (*Sebastodes* species) and the salmon (ocean Chinook or King, *Oncorhyncus tshawytscha*) steaks were all cut from raw fresh fish at the start of the experiments. The cooked Pacific

Table 1. Effect of beta radiation on the spoilage of sole fillets at 5.5° C (42° F) with various treatments.

Days stored	Volatile reducing substances (μeq/5 ml) in						Log bacterial numbers per ml press juice in					
	Control	CTC	IL	IH	CTC/IL	CTC/IH	Control	CTC	IL	IH	CTC/IL	CTC/IH
0	5.2	5.2	5.2	5.2	5.2	5.2	4.56	4.56	4.56	4.56	4.56	4.56
1	5.2	5.2	6.8	5.0	5.8	6.3	4.85	4.15	3.20	2.90	2.90	2.78
2	2.6	....	....	....	....	....	5.53	....	....	....	....	....
3	6.3	5.8	5.8	4.7	4.7	3.7	6.59	4.49	3.64	3.70	2.90	3.23
4	26.3	....	....	....	....	....	7.15	....	....	....	....	....
5	39.5	....	....	....	....	....	8.08	....	....	....	....	....
6		4.7	4.7	3.2	3.7	4.2	....	5.68	5.62	3.20	3.54	3.25
8		3.7	5.8	3.2	3.2	3.2	....	7.11	6.98	3.46	5.00	3.54
9		5.8	4.2	....	3.2	....	....	7.62	7.50	....	6.08	....
10		7.4	4.7	3.7	1.6	1.6	....	8.49	7.65	4.36	6.18	3.38
11		13.2	5.8	....	....	....	....	8.60	7.85	....	....	....
13		40.2	21.0	4.7	4.2	5.3	....	8.72	8.28	5.04	7.15	5.99
14			26.5	....	....	....	....	....	8.78	....	....	....
15				....	6.8	3.7	....	....	....	....	8.30	5.87
16				3.2	....	....	....	....	....	7.00	....	....
18				....	4.7	....	....	....	....	....	8.47	....
19				7.3	....	3.7	....	....	....	7.53	....	4.90
23				5.3	12.1	6.3	....	....	....	8.56	8.30	8.20
37				24.1		5.2	....	....	....	9.04	....	9.04

Days stored	Volatile nitrogen (mg/100 ml) in											
	Control		CTC		IL		IH		CTC/IL		CTC/IH	
	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN
0	0.6	0	0.6	0	0.6	0	0.6	0	0.6	0	0.6	0
1	0.6	0	0.6	0	0.6	0	1.9	0	1.9	0	1.9	0
2	0.9	0	....	....	....	....	....	....	....	....	....	....
3	1.2	0.9	1.6	0	1.2	0	0.9	0	1.6	0	0.9	0
4	3.7	2.5	....	....	....	....	....	....	....	....	....	....
5	10.5	5.6	....	....	....	....	....	....	....	....	....	....
6			1.6	0	1.6	0	1.6	0	0.6	0	1.6	0
8			3.8	0.3	1.9	0.6	1.9	0	1.9	0	2.5	0
9			1.9	0	2.5	0.6	....	....	1.9	0	....	....
10			1.9	0.6	1.9	0.3	1.9	0	1.5	0	0.9	0
11			3.8	1.2	3.1	0.6	....	....	....	....	....	....
13			10.1	4.7	4.4	3.1	1.9	0	1.9	0	1.9	0
14					10.8	6.3	....	....	....	....	....	....
15							....	....	3.7	0.6	2.5	....
16					13.1	8.1	5.6	0.3	....	....	....	....
18							....	....	4.6	0.6	....	....
19							5.2	0.6	....	....	3.3	....
23							3.9	0	5.3	2.6	5.3	0.6
37							11.8	4.9			6.6	0.6

All samples given a 5-minute dip in 5% NaCl solution. CTC—5 ppm chlortetracycline (CTC). IL—67,500 rads. IH—270,000 rads. CTC/IL—CTC + IL treatments. CTC/IH—CTC + IH treatments. Values with a broken line represent samples judged to be borderline passability. Underlined values represent samples sensorily judged spoiled.

Coast or Dungeness crabmeat *(Cancer magister)* and California shrimp *(Pandalus jordani)* were commercially frozen in hermetically sealed number 10 cans. They had been in frozen storage for several months and were thawed before use. The cleaned tuna loins were received precooked and frozen, and were thawed and cut into pieces before use. All the fillets and salmon steaks were packaged individually in polyethylene or polyethylene-polyester bags and heat-sealed. The shellfish were placed in polyethylene-polyester bags to yield a fairly flat package within the limits of penetrability of the source of radiation. The tuna loin pieces were placed individually in polyethylene bags and heat-sealed. All samples were kept cool by contact in a closed container with either wet ice or dry ice, but were not allowed to freeze. The untreated controls were treated similarly. They were taken to the radiation source in the same container, and then returned to the laboratory. The samples during the short irradiation periods, therefore, were all cold and at a temperature near 0°C.

Spoilage determination methods. As measures of the condition during storage, the contents of volatile reducing substances (VRS), total volatile nitrogen (TVN) and trimethylamine nitrogen (TMN) and total aerobic bacterial counts were determined on a press juice prepared from each sample by mechanical pressure, using nonabsorbent gauze and an aluminum cylinder and piston as a pressure chamber. The clean pressure chamber was treated with very hot tap water before the addition of the sample wrapped in the layers of gauze. The chemical procedures for the determination of VRS, TVN, and TMN have been described (Farber and Ferro, 1956). The total aerobic bacterial counts were made on suitable saline dilutions of the press juice. A 0.01-ml portion of three dilutions was spread on the marked-off surface of a Petri plate containing Difco Bacto Penassay Base Agar. Duplicate plates containing three dilutions were usually made for each press juice. The averages of the two determinations for each dilution were used. Each sample was also examined sensorily and its acceptability evaluated. In a few cases the samples were also steamed for 10 minutes and then similarly judged. Three laboratory personnel who were familiar with the products acted as the judges in all cases.

All the fish and shellfish samples were immersed for 5 minutes in a 5% NaCl solution. In some cases the appropriate amount of freshly prepared chlortetracycline solution was added to the salt solution before immersion of the fish or shellfish. All samples after immersion were drained for about 5 minutes and then packaged. The packages were stored at 30°F until transported to the source of radiation.

For some of the salmon steak experiments the immersion liquid also contained an emulsified antioxidant preparation containing known and recommended antioxidants. This antioxidant mixture was used in order to prevent as completely as possible the development of rancidity that takes place during storage even without any irradiation treatment.

RESULTS

Bottomfish. Low-fat fish. Tables 1 and 2 show the results of 2 separate irradiation experiments on sole fillets, some of which had been given an additional treatment with 5 ppm of chlortetracycline (CTC). It can be seen that the rate of spoilage of the fillets, compared to that of the controls, was definitely slowed down by both the antibiotic and the irradiation, and that a complementary action was found between the combined effects of the antibiotic and irradiation treatments. The lowest dose level of irradiation had an action similar to that of the 5 ppm of CTC—prolongation of the keeping quality by about a week. The highest dose level of irradiation inhibited spoilage for about a three-week period. The combination of antibiotic with the lowest or intermediate dose levels had approximately the effect of the highest dose level of irradiation.

Table 2. Effect of beta radiation on the spoilage of sole fillets at 5.5° C (42° F) with various treatments

Days stored	Volatile reducing substances (μeq/5 ml) in				Log bacterial numbers per ml press juice in			
	Control	CTC	I	CTC/I	Control	CTC	I	CTC/I
0	1.5	1.5	1.5	1.5	5.00			
1	6.3	6.3	2.6	3.0	4.56	4.58	3.74	2.90
2	5.2	....	....	....	6.50	....	....	....
3	5.2	....	....	....	6.93	....	....	....
4	6.3	....	....	....	7.58	....	....	....
5	21.6	....	....	....	8.47	....	....	....
6	37.5	3.7	5.2	3.1	8.72	4.75	6.60	4.60
8	....	5.2	....	....	....	6.56	....	....
10	....	6.3	10.5	5.7	....	7.08	7.61	4.53
11	....	6.3	12.0	....	....	7.38	7.75	....
12	....	9.7	18.0	....		8.00	7.80	....
13		35.3	31.7	6.7		8.34	7.72	7.49
16				2.6				7.64
17				8.4				8.04
21				27.7				8.50

All fillets were given a 5-minute dip in 5% NaCl solution. CTC—5 ppm chlortetracycline (CTC). I—Irradiated with 135,000 rads. CTC/I—CTC + I. Underlined values represent samples sensorily judged spoiled.

Medium-fatty fish. Irradiation of the somewhat fatty rockfish fillets with and without an additional CTC treatment, had results as shown in Table 3. In this case there was the added complicating factor of the unstable nature of the fish fat. When bacterial spoilage set in rather quickly, rancidification did not have any predominant part in the spoilage pattern. When bacterial spoilage was inhibited, however, by irradiation alone or in combination with CTC, some of the fillets became rancid before any bacterial spoilage, with the usual unpleasant decomposition odors, became evident. As a result, the efficacy of the irradiation treatment, either alone or with CTC, was not as marked as in the case of the low-fat flatfish. The prevention or slowing of rancidification by antioxidants would be expected to produce more marked inhibitory effect of the spoilage process.

Fatty fish. Salmon. The aforementioned possible added protection against rancidification by antioxidants was established in a series of tests on salmon steaks. The fish were treated with a composite mixture of known or recommended antioxidants in a concentration such as to expect good protection against the development of rancidity. The purpose, which was attained, was to eliminate or minimize the complicating fat spoilage. No attempt was made to determine and use minimally effective antioxidant levels. This aspect of spoilage prevention will be dealt with in a subsequent report. As in the case of the sole and rockfish fillets, spoilage of the salmon steaks was inhibited for about a week by 20 ppm CTC, and for about 4–5 weeks by rather low doses of beta irradiation (Table 4). The best effect of the combined treatments was found with the higher dose of irradiation complemented by the antibiotic. A further complicating factor noted in this series of tests was the development of extraneous irradiation odors in the film

Table 3. Effect of beta radiation on the spoilage of rockfish fillets at 5.5° C (42° F) with various treatments.

Days stored	Volatile reducing substances (μeq/5 ml) in				Volatile nitrogen (mg/100 ml) in								Log of bacterial numbers per ml press juice in			
					Control		CTC		I		CTC/I					
	Control	CTC	I	CTC/I	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	Control	CTC	I	CTC/I
0	9.0	9.0	9.0	9.0	0.3	0	0	0	0.3	0	0.3	0	5.40	5.40	5.40	5.40
1	6.9	6.2	5.1	5.1	0.3	0	0	0	0.3	0	0	0	5.53	4.60	3.66	3.76
2	10.6	....	4.8	....	4.0	0	....	....	2.6	0	....	....	6.30	....	4.66	....
3	15.4[d]	....	....	....	12.5	4.6	....	....	....	....	....	....	6.90	....	....	....
6	34.2[d]	6.1	2.2	3.3	9.2	5.8	2.0	0	0.6	0	3.3	0	7.61	6.78	5.69	5.88
7	....	3.9	5.7	4.4			1.6	0	2.6	0	3.3	0	....	6.83	5.59	5.48
8	....	4.4	....	....			4.9	0	....	....	....		....	7.25	....	....
9	....	6.6	14.5[a]	2.2			3.9	0	3.9	0.1	3.3	0	....	8.18	6.95	6.46
10	....	22.0[d]	7.5[b]	....			....	....	3.8	0.3	....	0		....	....	
11	....	20.0[d]	9.0[b]	9.7[b]			23.0	9.0	8.5	0.3	5.8	0		....		
13	....	39.1[d]	11.8[c]	12.1[c]					10.5	2.0	11.2	1.3		8.58	7.70	7.55
15	....	....	36.5[d]	29.6[d]					19.1	15.1	15.8	3.9		....	8.34	8.82
16	....	....	29.9[d]	18.9[d]					20.0	9.9	5.9	2.0			8.10	8.05

All fillets were immersed 5 min in a 5% NaCl solution. CTC—5 ppm CTC solution. I—125,000 rads. CTC/I—CTC + I.
[a] Rancid odor present—not considered acceptable.
[b] Very slight rancidity present—considered acceptable.
[c] Definite rancid odor present—less than in a.
[d] Definite spoilage odors present.

Table 4. Effect of beta radiation on the spoilage of salmon steaks at 1.7° C (35° F) with various treatments.

Days stored	Volatile reducing substances in							Log bacterial numbers per ml in						
	Control 1	Control 2	CTC	IL	IH	CTC/IL	CTC/IH	Control 1	Control 2	CTC	IL	IH	CTC/IL	CTC/IH
0	5.0	5.0	5.0	5.0	5.0	5.0	5.0	6.08	6.08	....	....	....	5.30	....
1	....	....	3.5	....	....	....	....	....	....	5.30	4.72	5.46	4.48	4.15
3	18.9[e]	7.5	....	....	....	....	....	7.48	6.81	....	....	....	....	
6	44.8	43.2[f]	....	....	....	....	....	7.15	7.81	....	....	....	....	
7	49.1	43.2	13.5	13.5	8.6	10.8	8.6	TNC	8.08	7.68	7.20	7.28	5.08	4.41
10	....	....	9.9	15.1	9.7	3.2	8.1			....	....	....	....	....
13	....	....	16.4[g]	5.9	5.9	1.0	0.5			8.23	7.48	7.57	7.00	6.68
15	....	....	....	14.1	....	....	....				7.50	....	....	....
17	....	....	....	12.9	9.1	4.8	9.1				7.86	7.56	7.69	7.41
24	....	....	....	....	15.6[g]	....	....				....	7.30	....	8.50
41	....	....	....	36.1	24.3	35.1	18.9[h]				7.38	8.08	9.21	8.45

Days stored	Volatile nitrogen (mg/100 ml) in													
	Control 1		Control 2		CTC		IL		IH		CTC/IL		CTC/IH	
	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN
0	....	....	....	....	....	....	....	....	....	....	....	....	....	....
1	....	....	....	....	....	....	....	....	....	....	....	....	....	....
3	12.5	1.3	6.6	0							....	....	....	....
6	7.9	1.3	6.6	0.6	....	....	....	....	....	....	....	....	....	....
7	16.5	7.2	14.5	6.6	6.6	0	6.5	3.3	3.9	0	2.6	0	3.9	0
10	11.7	3.2			5.8	0	9.1	4.5	6.5	0.6	6.5	0	6.5	0
13					15.1	7.8	6.6	0	7.2	0.6	2.6	0	1.3	0
15							7.2	1.3	....	....	....	....	....	....
17							4.6	0	4.6	0	1.9	0	5.9	0
24							....	....	4.6	1.3	....	....	13.2	0
41							6.6	1.9	5.9	0.6	5.9	1.9	6.6	0

All steaks were immersed about 30 minutes in 5% NaCl solution. Control 2—with antioxidant (AOX) emulsion. CTC—20 ppm CTC in AOX emulsion. IL—in AOX emulsion and 47,000 rads. IH—in AOX emulsion and 94,000 rads. CTC/IL—CTC + IL. CTC/IH—CTC + IH.

AOX mixture—final concn. in immersion liquids: Tenox VI—0.1%, NDGA—0.05%, THBP—0.05% (Prepared as a 10% stock solution in cottonseed oil); emulsifiers—2% TEM 4T and Tween 80

[e] Rancid, unpleasant odor.
[f] Spoiled odor, sharp and sour.
[g] Spoiled odor, good color.
[h] Some off odor present.
TNC—too numerous to count.

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bags, particularly after longer storage intervals. As a result, some of the high VRS values toward the end of the storage period likely reflected irradiation odors from the irradiated plastic film from which the packages were made. Nevertheless, a very definite extension in storage life was noted, as judged by chemical tests and sensory judgments of the raw steaks and after they were steamed for 10 minutes. The cooked taste of steaks receiving 94,000 rads was judged acceptable. Table 5 shows the chemical data for salmon steaks receiving doses of beta radiation varying from 11,800 to 94,000 rads. The increased inhibition of spoilage with increased dose level is evident.

The effect of a wide range of beta radiations on the stability of the color of the salmon steaks was also determined. Individual steaks were subjected to a series of dose levels of beta radiation starting with 23,500 rads and progressing up to 1,000,000 rads. As the dose level was increased from 94,000 to 305,000 rads a bleaching effect was observed, most pronounced at 610,000 rads: quite pale compared to the original color. On increasing the dose level to 10^6 rads another type of change apparently occurred, with the color darkening once more, but a deeper red instead of the original orange-red. There was a brownish component, resembling a flushed or feverish condition of the human skin.

Tuna. One orienting run was carried out on precooked tuna loins. The purpose was to determine whether the storage life of chilled precooked tuna loins could be significantly increased by irradiation. Table 6 shows the results. As noted many times, precooked tuna loins, unlike raw fish, do not spoil and develop strong unpleasant odors. In this case the control loins became moldy and unacceptable in about 25 days at 42°F, whereas precooked tuna loins that received 250,000 rads developed no detectable change other than some rancidity and yellow surface discoloration. Here again, an antioxidant treatment could have extended storage life still more.

Shellfish. Table 7 shows the prolongation of storage life and simultaneous inhibition of the spoilage rate of cooked Pacific crabmeat and California shrimp by beta irradiation doses of 50,000 and 250,000 rads. Crabmeat was apparently benefited more than shrimp, which nevertheless had an extension of almost 3 weeks with the combined treatment.

Table 5. Effect of varying doses of beta radiation on the spoilage of salmon steaks at 35° F (1.7° C) with various treatments.

Dose and treatment	VRS after days:							
	1	7	10	13	15	17	24	41
11,800, none	2.7	40.5	28.6	....	....	....	....	....
11,800 + 20 ppm CTC	2.1	5.9	1.6	14.0	13.5	....	....	....
23,500, none	2.7	32.4	29.7	....	....	....	....	....
+ 20 ppm CTC	2.7	3.2	1.6	0.5	....	5.9	....	....
47,000, none	3.7	13.5	15.1	5.9	14.1	12.9	....	36.1
+ 20 ppm CTC	3.7	10.8	3.2	1.0	....	4.8	....	35.1
94,000 none	2.1	8.6	9.7	5.9	....	9.1	15.6	24.3
+ 20 ppm CTC	2.7	8.6	8.1	0.5	....	9.1	....	18.9

Underlined values represent samples sensorily judged unacceptable or definitely spoiled.

Table 6. Effect of beta radiation on spoilage of steamed tuna loins at 42° F (5.5° C).

Days stored	Volatile reducing substances (μeq/5 ml) in		Log bacterial numbers per ml press juice in	
	Untreated	Irradiated with 250,000 rads	Untreated	Irradiated with 250,000 rads
0	4.3	6.4	4.78	5.48
7	2.7	3.7	5.96	5.59
25	5.4[j]	6.4	8.36	5.00
32	....	6.0	....	6.28
43	....	5.0[k]	....	6.92

[j] Loins moldy with musty odor; no longer acceptable.
[k] Some surface rancidity present.

Microbial sensitivity. Some experiments were carried out on the effect of various doses of beta radiation on the viability of representative microorganisms isolated from fresh and spoiled fish. In the first run, nutrient broth suspensions of *Pseudomonas* and *Achromobacter* strains from fresh and spoiled fillets of sole were found to be destroyed by 100,000 rads of beta radiation. In contrast, a strain of *Achromobacter* from spoiled fillets of sole that had been irradiated with 280,000 rads required 400,000 rads for complete destruction. In the second run, saline suspensions of *Flavobacterium* and *Pseudomonas* strains from spoiled fillets of sole were rendered nonviable by 25,000 rads, whereas similar suspensions of *Micrococcus* and *Achromobacter* from spoiled fillets of sole respectively required 75,000 and 50,000 rads for complete destruction. It was also found that irradiation of distilled water, saline, and nutrient broth with 3,200,000 rads markedly inhibited the growth of a strain of *Pseudomonas fluorescens* subsequently inoculated into these media and incubated at 25°C.

DISCUSSION

From examination of the above data a number of observations can be made. Low doses of beta radiation, as low as 11,800 rads, have a definite though limited inhibitory action on spoilage rate, which inhibition is further increased by chlortetracycline. Beta radiation in the range of 50,000 to 200,000 or 300,000 rads in a variety of fish and shellfish slowed up the onset of spoilage sufficiently to make practical applications possible, including a more widespread distribution of the treated products throughout the country.

In the case of medium-fatty fish, such as the rockfish species of the Sebastodes genus (which includes the commercially important Ocean Perch and Pacific Ocean perch varieties), and the definitely fatty fish, such as salmon, and tuna, fat spoilage during storage became a complicating factor in addition to protein spoilage, which generally becomes important only during extended storage periods at relatively high temperatures in the range of 30–40°F or higher. Significant preservation of these fish requires a suitable antioxidant to minimize or prevent fatty changes leading to rancidification. The effect of a lack of antioxidant became evident in the rockfish fillets and in the controls of the salmon experiments. The deleteri-

Table 7. Effect of beta radiation on spoilage of cooked crabmeat and shrimp at 35° F (1.7° C) after various treatments.

Cooked Crabmeat

Days stored	Volatile reducing substances (μeq/5 ml) in						Log bacterial numbers per ml press juice in					
	Control	CTC	IL	IH	CTC/IL	CTC/IH	Control	CTC	IL	IH	CTC/IL	CTC/IH
0	5.6	7.2	6.5	7.5	7.6	8.0	5.90	5.49	5.48	4.20	4.12	4.23
3	6.2	....	....	....	....	....	7.86	....				
4	9.5	....	....	....	....	....	8.02	....				
6	35.3	10.4	13.0	4.1	8.3	6.2	8.49	5.96	7.61	4.38	4.00	4.18
8	....	8.3	26.5	....	5.2	....	....	6.64	7.46	....	4.48	....
10	....	7.8	....	....	....	....	....	6.79	....	....	....	....
12	....	15.6	....	....	....	....	....	7.67	....	....	....	....
14	....	33.8	....	19.2	26.0	5.2	....	7.99	....	7.18	8.04	4.00
18	....	....	....	31.2	....	....	....	....	....	7.61	....	....
21	....	....	....	33.7	....	5.6	....	....	....	7.52	....	5.68
23	....	....	....	36.4	....	8.8	....	....	....	7.56	....	7.64
24						3.6	....	....	....	....	....	7.75
27						4.6	....	....	....	....	....	7.87

Days stored	Volatile nitrogen (mg nitrogen/100 ml) in											
	Control		CTC		IL		IH		CTC/IL		CTC/IH	
	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN
0	4.7	1.3	6.2	1.1	5.4	1.3	5.8	1.3	5.6	1.3	4.9	1.3
3	5.0	1.7	....	....	....	....	....	....	....	....	....	....
4	5.9	3.3	....	....	....	....	....	....	....	....	....	....
6	15.1	9.3	8.1	4.1	8.7	5.2	9.3	1.7	7.6	2.3	7.0	1.2
8			7.5	2.9	17.4	5.8	....	....	7.5	0.6	....	....
10			8.7	1.1	....	....	....	....	....	....	....	....
12			8.7	2.9	....	....	....	....	....	....	....	....
14			17.4	8.7	....	....	11.6	2.9	17.4	3.4	5.8	0.6
18					....	....	18.5	4.6	....	....	....	....
21							14.0	8.5			5.0	0.6
23							17.4	7.7			9.8	1.1
24											5.8	0.6
27											13.9	1.7

Cooked Shrimp

Days stored	Volatile reducing substances (μeq/5 ml) in						Log bacterial numbers per ml press juice in					
	Control	CTC	IL	IH	CTC/IL	CTC/IH	Control	CTC	IL	IH	CTC/IL	CTC/IH
0	8.7	6.5	6.7	7.0	8.7	8.2	5.89	5.49	4.48	4.62	4.90	4.20
3	9.2	....	....	....	....	....	7.89	....	....	....	....	....
4	14.5	....	....	....	....	....	7.90	....	....	....	....	....
6	37.4	8.3	18.2	6.7	11.4	5.2	7.90	7.46	7.86	3.96	5.92	3.30
8	....	29.6	31.7	....	9.3	....	....	7.90	7.83	....	6.57	....
10	....	39.0	....	....	....	....	....	8.12	....	....	....	....
14	....	....	....	6.7	38.4	7.8	....	....	....	7.18	7.51	7.46
19	....	....	....	9.2	....	....	....	....	....	7.30	....	....
21	....	....	....	18.2	....	8.5	....	....	....	7.85	....	7.76
23	....	....	....	39.0	....	23.4	....	....	....	8.43	....	7.69

Days stored	Volatile nitrogen (mg nitrogen/100 ml) in											
	Control		CTC		IL		IH		CTC/IL		CTC/IH	
	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN	TVN	TMN
0	4.8	2.0	5.5	1.3	6.0	1.3	6.0	1.3	4.2	1.3	4.1	1.3
3	5.1	2.3			....	....	....	....	....	....	....	....
4	5.2	....	....	....	....	....	....	....	....	....	....	....
6	14.5	11.2	8.7	1.7	9.3	5.2	6.4	1.7	11.1	5.2	8.7	1.2
8			16.8	4.9	17.4	6.3		....	8.7	2.9		
10			20.3	14.5	....	....	....	....	....	....	....	....
14							8.7	1.1	41.1	13.8	12.1	1.1
19							7.8	3.1	....	....	....	....
21							8.3	6.0	....	....	6.1	1.3
23							24.9	16.3			11.6	4.6

All the shellfish were immersed 5 min in a 5% NaCl solution. CTC—25 ppm CTC in 5% NaCl solution. IL—50,000 rads. IH—250,000 rads. CTC/IL—CTC + IL. CTC/ IH—CTC + IH. Underlined values represent samples sensorily judged unacceptable or spoiled.

ous action of beta rays on fish fat was pointed out by Astrack *et al.* (1952) and by others (Liston and Shewan, 1957; Nickerson *et al.*, 1950; Symposium, 1954). The efficacy of an antioxidant for significant prolongation of the storage life of salmon is also evident from the above data.

Lukton and Mackinney (1956) showed that doses of gamma radiation on the order of 250,000–1,000,000 rads produced a bleaching of the color of salmon oil, and that 1,000,000–4,000,000 rads caused a progressive bleaching of the color of boiled prawns. The bleaching effect on salmon color was observed in our experiments too. No antioxidant was present in these samples, since the effect of the beta radiation alone was being studied. It is possible that an antioxidant could decrease or inhibit the decolorization of salmon at these higher dose levels. This possibility will be determined in our later studies on the effects of beta radiation.

Anderson *et al.* (1956) described a micrococcus that became resistant to 6 million rep of gamma radiation. From fillets of sole irradiated with 280,000 rads that ultimately spoiled we isolated an *Achromobacter* strain that was somewhat more resistant to irradiation than similar organisms isolated from fresh fish or from unirradiated fish that had spoiled. The relative sensitivity of *Flavobacterium*, *Pseudomonas*, and *Achromobacter* species to relatively low doses of either beta or gamma radiation has been mentioned a number of times (Goldblith *et al.*, 1953; Hannan, 1955; Lück and Kuhn, 1959; MacLean and Welander, 1960; Nickerson *et al.*, 1950; Niven and Chesbro, 1957). These experiments similarly showed the sensitivity of strains of these genera to relatively low doses of beta radiation, as well as the somewhat greater resistance of strains of micrococcus, which also has been described previously (Hannan, 1955).

Similar to the fish species already mentioned, cooked Pacific crabmeat (Dungeness crab) and California shrimp, when treated with beta radiation, were also markedly preserved, the increased storage time being dependent on the dose level. No bleaching of the color of the cooked shrimp was noted in these experiments with 50,000 and 250,000 rads, in contrast to the aforementioned observations of Lukton and Mackinney (1956), who started with 1 million rads of gamma radiation and found a progressive bleaching of the color with increasing dose levels.

It has been pointed out many times (Anon., 1954; Hannan, 1955; Huber, 1951; Lück and Kuhn, 1959; Symposium, 1954) that the preservative effect of beta and gamma radiation may be in part the result of their action on the suspending medium or menstruum, with the production of free radicals that exert a destructive action on the constituents of living organisms or of foodstuffs, such as fat. Mention has been made of a preliminary run on the effect of a high dose of beta radiation on three media, and the subsequent inhibitory action of the irradiated media on a strain of *Pseudomonas fluorescens* suspended in them. It is evident that the irradiated media had a destructive or inhibitive action on the organism. Further work is also planned on this aspect of irradiation.

The complementary action of relatively low doses of beta radiation and antibiotics of the tetracycline class, as exemplified by chlortetracycline, was evident for all the types of fish and shellfish studied. For the fatty fish, such as salmon, the triple combination of antioxidant–antibiotic–low dose of beta radiation was quite effective in significantly extending storage life at refrigerated temperatures above the freezing point. With this combination of complementary treatments, country-wide distribution of fresh chilled but unfrozen salmon becomes a possibility. A similar greater distribution pattern would also be feasible for the other commercially important Pacific Coast raw fish.

One requirement remaining to be completely fulfilled is an odorless packaging film that will remain so after irradiation with the doses of beta radiation used in this work. As mentioned, volatile material present in the packages in some of the runs was reflected by an increase in VRS content even though the product was sensorily judged to be still acceptable.

Acknowledgments

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Evaluation of New Strawberry Varieties for Freezing and Preserving[a]

(Manuscript received July, 1960)

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Fruit and Vegetable Products Laboratory[b] and Western Washington Experiment Station, Puyallup, Washington

SUMMARY

New strawberry varieties were compared with Marshall, the long-time standard variety for freezing and preserving. Varietal and seasonal differences were noted in ascorbic acid, titratable acidity, soluble solids, and solids-to-acid ratio. The trend among varieties was to fall in similar order in each of the seasons covered, though variations were wide in composition and in quality as evaluated by taste panel among samples of the same variety grown in different areas.

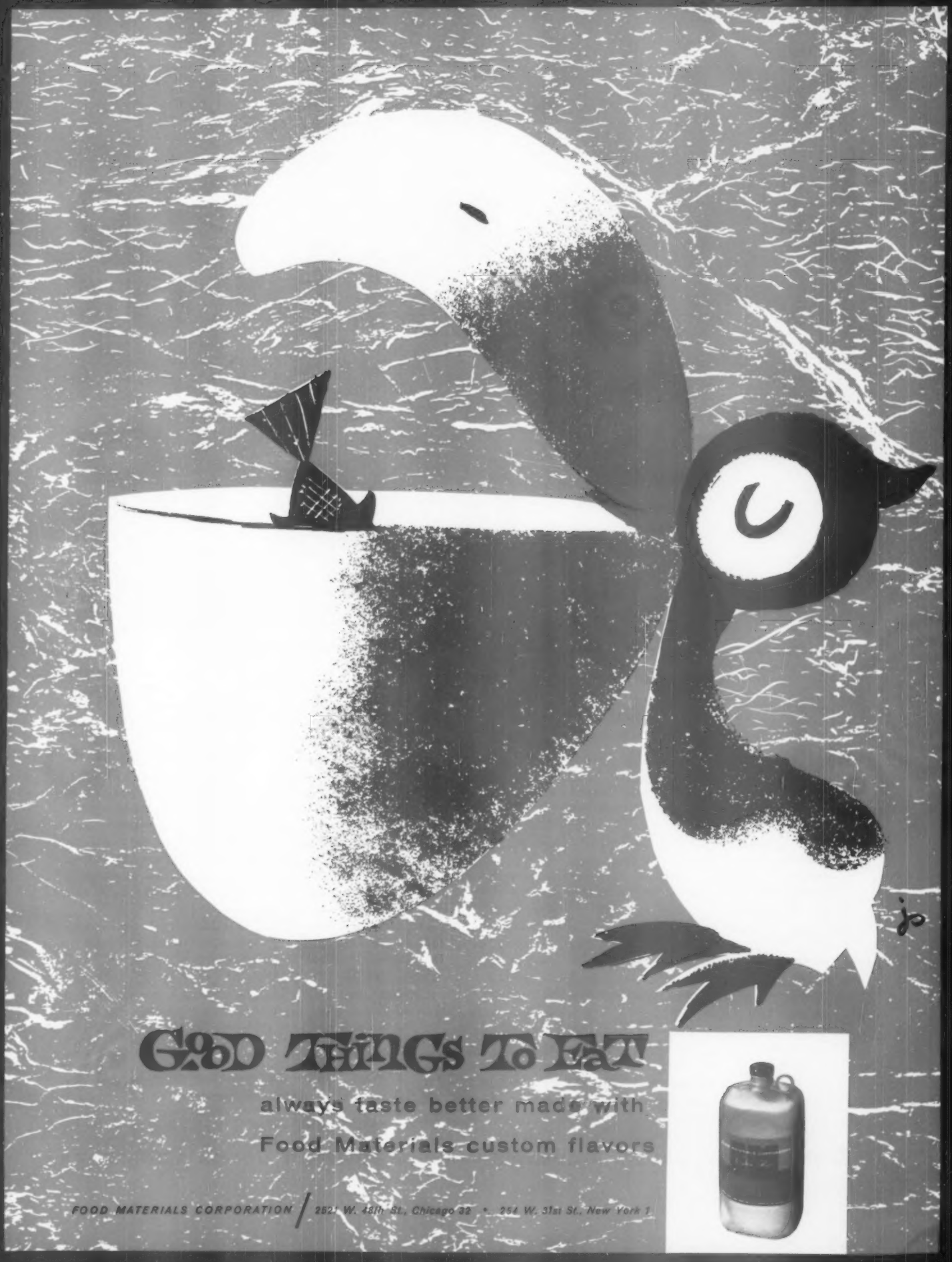
On the basis of flavor, wholeness of fruit, and color stability, some of the new selections compared favorably with the Marshall variety for preserving.

THE STRAWBERRY variety picture changes slowly and constantly. Older varieties lose ground as they fall victim to degenerative diseases or as superior varieties are developed. For example, in 1937, according to Darrow and Waldo (1939), the leading processing varieties grown in western Washington were Marshall, Corvallis, Ettersburg 121, and Redheart. Ten years later the same authors rated Marshall, Brightmore, Corvallis, and Redheart (Darrow and Waldo, 1948) as the leading varieties grown there. In 1958 Dodge and Snyder (1959) noted that the leading varieties were Northwest and Marshall, along with small acreages of Siletz and Puget Beauty. Of varieties presently grown, only Marshall existed as a named variety prior to 1949.

Breeding of new strawberries is being vigorously pursued by Washington State University (WSU) at the Western Washington Experiment Station, and by Oregon State College (OSC). The project in Oregon is conducted in cooperation with the U. S. Department of Agriculture. Promising hybrids selected by the plant breeders are being tested for processing quality at OSC by the Food Technology Department and in Washington by the USDA Fruit and Vegetable Products Laboratory. This paper deals with the evaluation of strawberry varieties in the latter labora-

[a] Presented at the annual meeting of Institute of Food Technologists, San Francisco, May, 1960. Scientific Paper No. 1968, Washington State University Agricultural Experiment Station Series.

[b] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.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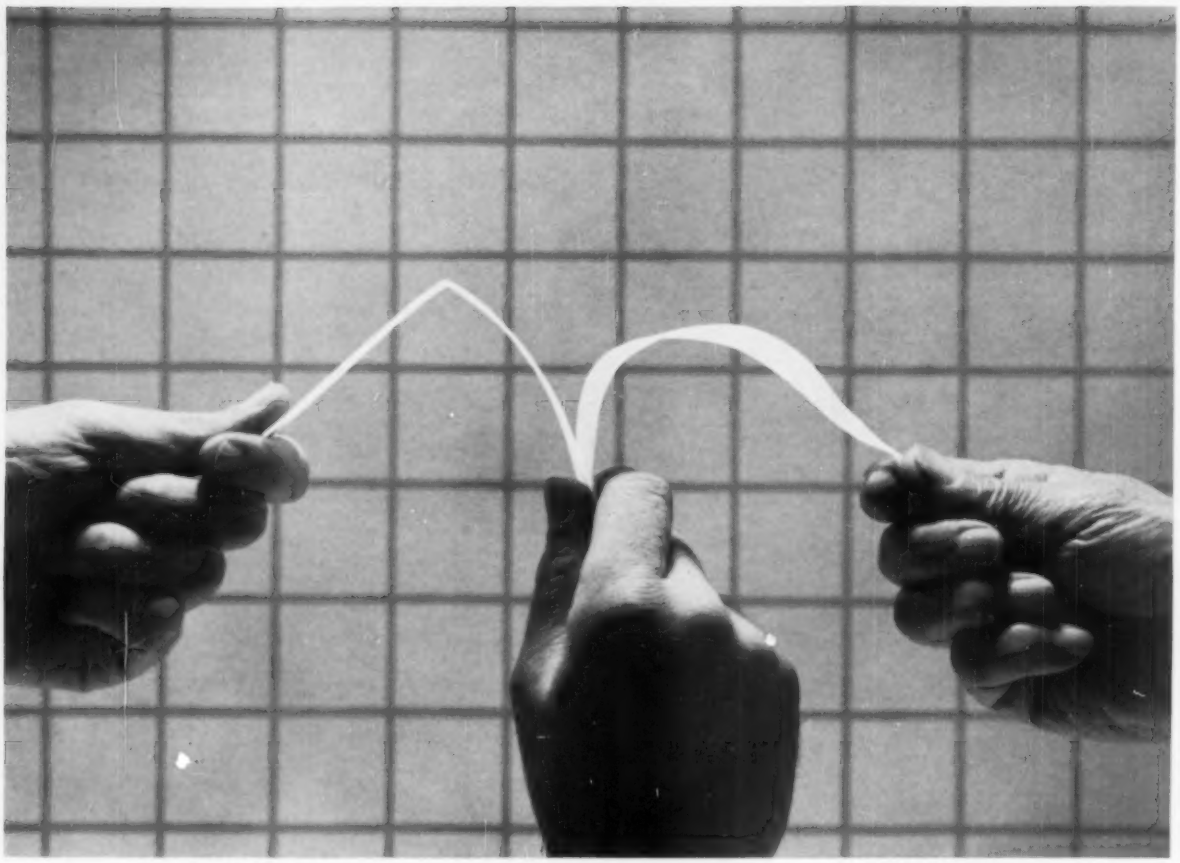

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tory. In particular it deals with 1957 and 1958 work in which the long-time standard processing variety Marshall was compared with Northwest, introduced by WSU in 1949; Siletz, released by OSC-USDA in 1956; Puget Beauty, introduced by WSU in 1956; and Cascade and Columbia, released by WSU in early 1960.

The first evaluation of new hybrid strawberries is done by the plant breeder. He propagates those most promising and puts them through a series of field tests of yield, disease resistance, ease of picking, and handling quality. He discards all that are noticeably deficient in size, color, texture, or flavor. Only a small number of seedlings are ever carried far enough along for processing studies. Of several thousand seedlings coming from a series of crosses, only a dozen or so may show enough promise on the basis of growth characteristics, disease resistance, production, and fruiting habits to warrant their being propagated in quantities large enough to permit freezing and preserving studies. Experimental plantings of these selections are usually grown in several districts, such as Puyallup, Vancouver, and Mount Vernon, to determine their adaptability to soil and climate variation.

In studies to be reported, daily contacts were maintained with the harvest crews so that the day's processing activities could be planned. Several trips a day were made to the picking sheds in order to select the fruit needed for the studies. Randomized selection of filled flats from each variety harvested was made to be sure that berries obtained represented the entire picking, not just berries from a single row. After the preliminary cleanup picking made in the Experiment Station plots, every picking was sampled for chemical testing. The freezing of berries for taste-panel evaluation was usually confined to the second and third pickings. Later pickings, consisting of smaller berries, were bulk-packed for preserving studies.

Berries from Vancouver and Mount Vernon plots were selected from the morning pickings and brought to the laboratory by automobile. Approximately 3 hours' transport time was involved with berries from these two stations, 135 miles south and 100 miles north of Puyallup. Only the second and third pickings were sampled from these two locations.

PROCESSING

The berries were all processed on an experimental line similar to production lines in several processing plants in the area. This line consisted of a shaker washer, an inspection belt, a size grader, and a power-driven slicer. In freezing tests 400 g of sliced berries were weighed into No. 2 dark-fruit enameled cans. A predetermined amount of sugar based on the soluble solids of the sliced fruit was added to bring the solids content of the sweetened berries to 25%. The cans were sealed and the sugar and berries mixed by inverting each case of cans 30 times at 30-second intervals. They were then blast-frozen at −20°F and stored at −5°F.

Four plus one berries for preserving studies were packed by filling 30-lb enameled tins with 24 lb of washed berries and 6 lb of sugar. The filled cans were lidded and placed at −5°F for freezing and storage.

Preserving Tests. Frozen 4 + 1 strawberries were thawed by holding the 30-lb tins in a bath of cool water overnight prior to making them into preserves. A standard 45% fruit to 55% sugar by weight formula was used, and the cooking followed a standardized procedure as follows: Drain the syrup from the thawed berries and add it to the pectin solution in the steam kettle used for cooking the preserves. One quarter ounce of 150 grade rapid-set pectin was used for each 10 lb of berries. Cook the syrup until 216°F is reached, add berries, heat to 210–212°F, and add sugar. Cook to 223° or 68% soluble solids, add citric acid equal to 1 g acid per pound of fruit. Cool quickly to 190° and fill into jars. Seal and cool by immersing the jars in warm, then cold, water. After filling and cooling, control jars of each lot of preserves were stored at 34°F. Test samples were stored at higher temperatures, including 70 and 98°F. Control material was used for comparisons of color of preserves stored at higher temperatures. Flavor comparison was made by pairs.

Chemical Analysis. A 5-lb composited sample of washed berries was taken from each lot of fruit for analysis. This sample was thoroughly mixed, and a full blender jar taken for analyses. Samples were blended for about 1 min. The soluble-solids content of wet fruit was measured on the Abbé refractometer, and duplicate 25-g portions were taken for total-acidity and ascorbic-acid determinations.

Titratable acidity was determined by titrating to pH 8.1 with 0.1*N* NaOH. The acid content was expressed as grams of anhydrous citric acid per 100 g. The soluble solids–acid ratio was determined by dividing the per cent soluble solids by the per cent citric acid found in the sample. Reduced ascorbic acid was determined by the method of Loeffler and Ponting (1942).

Appraisal. A panel of six experienced judges evaluated the frozen, sliced berry pack. Before any intervarietal comparisons were made, a sample of Marshall strawberries was defrosted, cut, and rated for color, appearance, flavor, and texture. Each factor was discussed, and agreement was reached by the panel as to the scoring of the sample. In subsequent sessions, when no discussions were allowed during the tasting, a labeled sample of the same lot of Marshall was presented for tasting along with five unknown samples. The unknowns were compared with the previously pegged Marshall standard in making intervarietal comparisons. Occasionally, a check sample of the standard was included in the five unknown samples. In all tasting, a No. 2 can was considered a sample. The thawed berries were emptied into an enameled pan for color and appearance factors. After these factors were scored, each taster removed 50–75 g of the berries for scoring of flavor and texture.

Grading was on a 1–10 basis: 10, excellent; 7–9, good; 5–6, fair; 3–4, poor; and 1–2, very poor. A top rating for color was given to berries having a uniform, bright, live-red color. Dull, dark color was rated down, as were pale, "washed out" colors. Factors considered in appearance were the degree to which the identity of the slices was maintained and the amount of collapse or breaking up of the slices. Desirable texture is best described as fresh-berry "feel" in the mouth. Top-rated berries were neither soft and slippery nor rubbery, but had firmness without toughness. Flavor was more difficult to assess, but the desirable features looked for were a pleasing balance between acidity and sweetness, with aromatic characteristics that were strawberry-like and did not remind the taster of other fruits.

RESULTS AND DISCUSSIONS

All tests reported in Table 1 were made on washed fruit. A separate investigation was made of the effect that washing would have on the solids and ascorbic acid content. Dry fruit averaged 0.4% higher in soluble solids as measured by refractive index than washed berries, but the ascorbic acid content of the dry fruit was slightly lower. The results have not been corrected for lowering of soluble solids content by washing. In Table 1, the average values for soluble solids, per cent acid, soluble-solids-to-acid ratio, and mg per cent ascorbic acid are given. The figures

Table 1. Soluble solids, per cent acid, soluble solids–acid ratio, and ascorbic acid contents of washed fresh strawberries during 1957-1958 seasons

Variety	Soluble solids		Per cent acid[a]		Ss/acid ratio		Mg per cent ascorbic acid	
	1957	1958	1957	1958	1957	1958	1957	1958
Marshall	6.7 (5.8–7.4)	6.5 (5.4–7.7)	.69 (.61–.75)	.77 (.65–.92)	9.8 (8.6–10.8)	8.1 (7.6–9.1)	50.0 (40–71)	53.8 (30–77)
Northwest	7.0 (6.0–7.6)	6.3 (5.2–7.4)	.86 (.75–.94)	.80 (.68–.94)	8.2 (7.8–8.9)	7.9 (6.6–9.0)	55.0 (47–61)	47.5 (28–63)
Puget Beauty	9.2 (8.4–11.5)	7.6 (6.1–10.3)	.86 (.67–1.03)	.76 (.63–.83)	11.0 (9.7–13.6)	10.1 (8.6–16.4)	58.0 (46–67)	51.5 (34–79)
Siletz	7.9 (7.3–8.8)	6.1 (4.9–7.4)	.93 (.87–.96)	.79 (.67–.91)	8.4 (7.8–9.1)	7.3 (6.5–10.1)	49.4 (44–54)	39.9 (25–47)
Cascade	8.3 (8.2–8.4)	6.7 (5.5–7.8)	.88 (.85–.90)	.83 (.74–102)	9.9 (9.6–10.0)	8.2 (6.6–9.6)	44.5 (41–48)	31.0 (28–45)
Columbia	8.3 (8.0–8.8)	6.5 (5.5–7.5)	.96 (.90–1.01)	.89 (.71–1.16)	8.6 (8.0–9.0)	7.4 (6.4–9.4)	61.0 (59–68)	55.0 (39–92)

[a] Per cent acid expressed as anhydrous citric acid.

in parentheses immediately below each average value denote the high and low values found during the season for the variety.

Except for the total acid and ascorbic acid content of Marshall, all constituent values listed in Table 1 were lower in 1958 than in 1957. The reason is not known, but weather may have had a part in it. The period (May, 1958) when the berries were developing was unseasonably warm for the area.

Puget Beauty, a very sweet berry, had the highest soluble solids and soluble-solids-to-acid ratio during both seasons, whereas Siletz ranked 5th in 1957 and 6th in 1958 in soluble-solids-to-acid ratio. The Cascade strawberry was lowest and Columbia highest in average ascorbic acid content during both seasons. Puget Beauty and Marshall averaged 50 mg per cent or higher in both years. Northwest dropped below 50 mg per cent in 1958, and Siletz was less than 50 mg per cent in both seasons.

Table 2 shows the results of the analysis of strawberries grown in 3 locations. The variation between samples of a variety as well as with location is given. These samples were all third-picking fruit.

There is a spread of season between locations, with Mount Vernon coming on a few days later than Puyallup and about a week later than Vancouver. The weather was unsettled in the Vancouver area when the berries were harvested, whereas at Mount Vernon good weather prevailed at the time the third picking was made. The higher ascorbic acid values in the berries from Mount Vernon may reflect the weather conditions in the location, since it has been shown that berries picked in prevailing sunshine tend to be high in ascorbic acid (Hansen and Waldo, 1944).

In taste-panel comparisons of the frozen berries (Table 3), all samples had good color in 1957, but only Marshall and Northwest maintained these high ratings in 1958. In appearance and texture, Marshall, Puget Beauty, and Siletz fall into a lower group, with Northwest, Columbia, and Cascade decidedly better in appearance and texture. Cascade was exceptional in texture after defrosting. The flavor factor is difficult to assay, since personal likes and dislikes are bound to enter into the ratings. Marshall and Cascade were well liked, because of a balance between sweetness and tartness and also because of their pleasant aromatic characteristics. Puget Beauty was too sweet to some tasters, who sometimes likened its aromatic character to that of apricots, plums, or apples.

Table 2. Analysis of washed fresh strawberries from different locations, 1958 season—single samples of third picking fruit from each location

Variety	Location	Per cent soluble solids	Per cent acid	ss/acid ratio	Mg % ascorbic acid
Northwest					
	Puyallup	6.0	.78	7.7	47
	Mt. Vernon	7.0	.83	8.6	63
	Vancouver	5.2	.68	7.7	50
Puget Beauty					
	Puyallup	6.7	.75	8.9	50
	Mt. Vernon	8.3	.81	10.2	57
	Vancouver	7.5	.88	8.6	34

Table 3. Varietal evaluation of frozen sliced strawberries by a panel of 6 experienced judges

Variety	Color		Appearance		Flavor		Texture	
	1957	1958	1957	1958	1957	1958	1957	1958
Marshall	8.0[a]	8.0	7.0	7.0	7.7	8.0	5.6	7.0
Northwest	8.8	9.0	8.6	8.2	7.0	8.1	8.1	7.9
Puget Beauty	8.1	7.0	7.9	6.0	8.0	7.0	7.9	5.8
Siletz	8.0	6.7	7.4	6.7	7.1	5.8	6.3	5.3
Cascade	8.9	7.2	8.8	8.5	8.5	7.5	8.8	8.7
Columbia	8.0	6.7	8.8	7.5	7.2	6.8	8.2	7.8

[a] Scores are averages of panel's ratings of 4–8 tastings of each variety each season. 10 = excellent; 7–9 = good to very good; 5–6 = fair; 4 and lower = poor.

Table 4 presents the panel evaluation of frozen berries packed from the same lots of berries for which soluble solids, etc., are given in Table 2. There were differences showing in all factors among the samples of the same variety. These differences were as great as those between varieties. In both varieties the samples having the highest solids-acid ratios were rated highest by the panel.

It was observed in the preserving work that Puget Beauty foamed more during cooking than other varieties. With every variety, floating of the berries in the hot syrup before it jelled was troublesome at times. The use of half berries helped control floating. With half berries, gases and steam could escape through the cut side of the berries, and there was also less break-up of berries during the cook. Because appearance plays an important part in preserve sales, it is important that color be stable. Neither a pale,

Table 4. Panel evaluation of frozen strawberries from different locations, 1958 season.[a] Single samples of third picking fruit from each location

Variety	Location	Color	Appearance	Flavor	Texture
Northwest					
	Puyallup	9.0	7.8	7.6	7.6
	Mt. Vernon	7.8	7.8	7.8	7.8
	Vancouver	6.8	6.2	6.6	7.0
Puget Beauty					
	Puyallup	8.0	7.2	7.5	7.3
	Mt. Vernon	7.8	8.5	8.2	7.5
	Vancouver	7.7	6.8	7.3	6.2

[a] These berries are from the same lots for which analyses are given in Table 2.

faded product nor a blackened preserve is desired. Cascade preserves had the slowest color change, which may be related to its lower ascorbic acid content. Sondheimer and Kertesz (1953) have presented evidence linking ascorbic acid to destruction of anthocyanin in strawberry juice.

When the preserve samples were examined for color change, the jars were placed in a strong beam of white light to illuminate the contents of the jar, showing up both the berries and the jelly. Using the 35°F samples as control and comparing within the variety, it was possible to follow color changes of the variety under different storage conditions. In the 50°F samples, a slight change in berry color, but not in the jelly, was noted in all varieties. At room temperature the change was quite noticeable in the berries in all preserves, but the Cascade sample; it showed little change over that observed in the sample stored at 50°F. All 98°F storage samples showed considerable fading of the berries. The jelly surrounding the Cascade berries was still a pleasing red, however. The above observations were made after 4 months' storage at the temperatures of the test.

It was our experience that it was difficult to maintain interest of the formal taste panel in getting flavor evaluation of preserves. Preferences were obtained, however, by issuing paired samples to the Experiment Station staff along with a form asking for a report on numbers of persons tasting and their preferences. On the basis of reports returned it was found that preserves made from the three varieties having pronounced aromatic characteristics (Cascade, Puget Beauty, and Marshall) were preferred by tasters, although none had a wide margin of preference. On the basis of flavor, color, and wholeness of berries, Cascade was found to be the best of the newer varieties.

On several occasions we have submitted preserve samples to manufacturers for their evaluation. The preservers' reaction to Cascade has generally been that samples were equal to, if not a little better than, those made from Marshall.

The present study is too limited in duration for correlation of chemical composition with freezing quality. Freezing quality is the term used by the authors to indicate the desirability of color, appearance, flavor, and texture of strawberries after freezing and defrosting. Soluble-solids content, pH, titratable acidity, solids-to-acid ratio, and ascorbic acid content may influence the quality of the strawberries but still not indicate how well the varieties will freeze. Neither do they indicate what the flavor will be beyond a suggestion as to whether the berries will be sweet or tart. Strohmaier and Pen (1954), in California, and Robinson *et al.* (1947), in New York State, also found it difficult to discover a relationship between composition and freezing quality.

Because the strawberry is subject to variation, evaluation of a variety on the basis of a single sample is very unrealistic. Varieties differ, but berries from the same variety may also differ widely in soluble-solids and acid makeup. To evaluate strawberry varieties for intelligent processing, berries from different locations should be studied for more than a single season. The sampling should be as frequent as possible during the season, and close contact with grower and harvest crews is advisable to ensure optimum control of the fruit being studied.

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The Color of Raw Tomato Juice*

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SUMMARY

Chromaticity coordinates x and y and luminous reflectance Y of tomato juice are not affected by cell depth of 10–150 mm. Length of holding time and deaeration treatment affect the color of raw tomato juice prepared from individual tomatoes. Data on the color range of five tomato varieties indicate that the chromaticity coordinate x and luminous reflectance Y are directly correlated with the maturity of the sample.

INTRODUCTION

It is generally recognized that color is an important factor in evaluating quality grades of tomatoes and tomato products. Many investigators have reported on tomato color with regard to various aspects of the grading problem (Desrosier, 1954; Kramer, 1954; Wilson and Dever, 1954; Younkin, 1957). In particular, the measurement of tomato color by visual (subjective) as well as photoelectric (objective) techniques has been described and correlated with visual ratings (Kramer, 1954; Robinson *et al.*, 1952). Photoelectric measurement of tomato color has been commonly found the most desirable aim in grading procedures (Yeatman and Sidwell, 1958). Tomato color has been evaluated photoelectrically in whole tomatoes, cut tomatoes, raw juice, and the various forms of processed tomatoes. A survey of the literature, however, shows that in studies on the color of raw tomato juice the interpretation of effects of holding time and deaeration treatment on the color of raw juice appear to be controversial. While, for example, Gould *et al.* (1954) and Hand *et al.* (1953) found it necessary to deaerate the juice under vacuum before color measurement, Yeatman and Sidwell (1958) reported that, although deaeration improved color, the "improvement was so constant and predictable that it was found logical to discontinue deaeration, thus simplifying the procedure."

The effect of variety on the color characteristics of a juice sample has also received little attention. In general the color of raw tomato juice is measured on a composite sample prepared from a mixture of different fruits from one or more varieties. Yeatman and Sidwell (1958), however, showed that such color measurement may be biased. In particular it was found that the resultant color of a blend was significantly biased toward the color of the redder sample.

In the present study the use of composite samples is avoided, and each color measurement is made on the juice derived from a single tomato. In the first part of this study the effects of holding time and deaeration treatment on the color of raw tomato juice from individual fruits at three stages of maturity have been evaluated. In the second section are presented data on the color range of raw tomato juice as determined by juice samples of individual fruits of five different varieties at three stages of maturity.

MATERIALS AND METHODS

1. *Effect of holding time and deaeration treatment on the color of raw tomato juice.*

For this part of the study the tomatoes used were var. Ferguson grown at the Central Experimental Farm at Ottawa. For each experiment four tomato fruits of each of three different stages of maturity were picked about one hour before use. The three stages of maturity were judged visually by exterior color and specified as light red, medium red, and dark red. All fruits were free of blemishes. Adherent soil and spray residue were washed off, and the calyx and blossom end were removed. The fruits were juiced individually by putting the whole or cut tomato into a small juicer (Fig. 1) placed in a Carver Laboratory hydraulic press. The juice so obtained was free of seeds and skins, and contained about 4% (by volume) air bubbles. The total solids content and color of juice so prepared were equal to those of juice prepared with

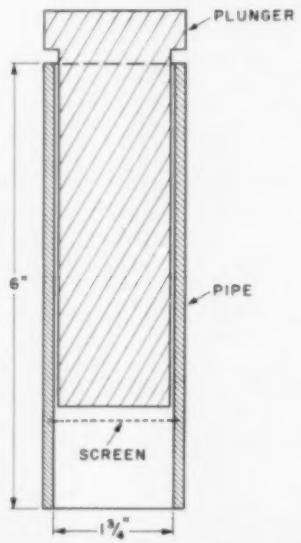
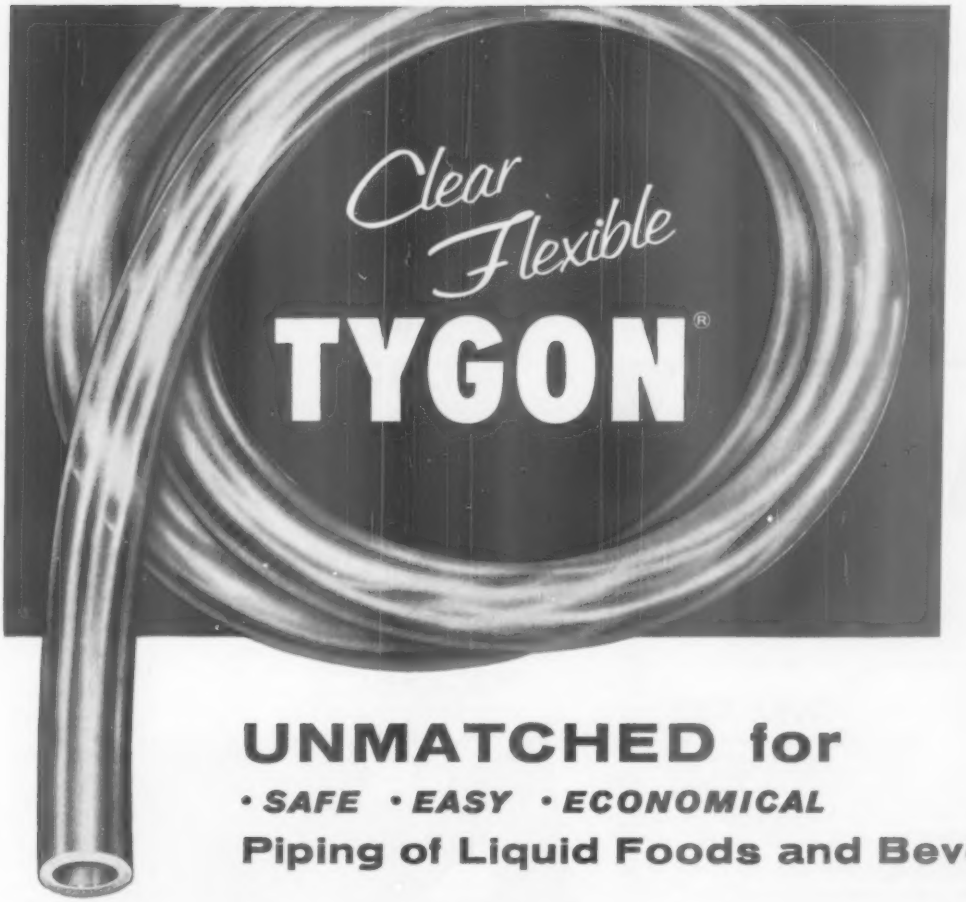


Fig. 1. Cross-section of juicer used to obtain juice from individual tomatoes. Plunger and pipe are made of brass, the screen is made of stainless steel, with 1/32-in. holes at 1/16-in. centers. The whole instrument is silver-plated (0.003-in.).

* Contribution No. 25, Plant Research Institute, Research Branch Canada Department of Agriculture, Ottawa, Ontario.



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the Langsenkamp Laboratory Juicer. The effect of holding time and deaeration treatment on the color of raw tomato juice was then studied by submitting each group of 12 juice samples to one of the following conditions: 1) untreated juice held at room temperature for 30, 60, and 90 minutes; 2) two drops of n-octyl alcohol added to each 100 ml of untreated juice and held at room temperature for 30, 60, and 90 minutes; and 3) untreated juice exposed to vacuum (15 repetitions of alternate drawing and breaking of the vacuum) and held at room temperature for 30, 60, and 90 minutes.

The effects of the treatments on the color of the juice samples were measured on a Hunter Color and Color Difference Meter in conjunction with a calibrated (National Bureau of Standards) porcelain enamel standard with Hunter coordinates L = 25.6, a = 28.3, b = 13.6. The performance of the instrument was checked, every third sample, by measuring another calibrated porcelain enamel standard (L = 24.2, a = 19.8, b = 5.1) against the first one. The differences of the color coordinates were obtained by subtracting the coordinates of the treated sample from those of the original sample.

The instrument mirrors were in the "s" position, and the 1-in. aperture plate was used for all measurements; the juice samples were submitted in a cylindrical quartz container (2½ diameter), filled to a depth of 1¾ in.

2. *The color range of raw tomato juice.*

At three dates (Sept. 4, Sept. 10, Sept. 18) during the 1958 season, fruits of different varieties and maturities were collected for determination of the color range of raw tomato juice. The following varieties were chosen: Ferguson, John Baer, San Marzano, Webb Special, and UI 30. On each occasion the juice of each individual fruit was prepared as described above and filled into a quartz cell (2.5 × 2 × 0.5 in.) to measure its spectral reflectance on a General Electric Recording Spectrophotometer with an attached digital tristimulus integrator (White and MacAdam, 1957). A piece of black velvet was placed around the cell, leaving open only the front, which was held against the sample port of the integrating sphere of the spectrophotometer. The measurements were made relative to magnesium oxide. The specular component of the sample was excluded. It took about 5 min to complete one run over the spectral range of 400–750 mμ. Simultaneously the C.I.E. tristimulus values X, Y, Z were obtained on the digital tristimulus integrator. Simple calculations converted the X, Y, Z values into C.I.E. x,y chromaticity coordinates. This procedure of measurement closely follows that used by Younkin (1957) for measurements on tomato purees.

On the basis of some measurements repeated on the same juice samples, but stirred before remeasured, it was observed that the chromaticity coordinates x and y and the luminous reflectance Y could respectively be repeated within ±0.002, ±0.002, and ±0.2. These values are in each case about twice as large as those usually obtained for colored samples with a smooth and flat surface, such as paint samples or glass filters.

Some thought was given to the concept of the spectral reflectance of a translucent material such as tomato juice. McCollum (1944) and Robinson *et al.* (1952) pointed out that the reflectance measurement should be independent of the type of cell backing. Whenever a change in reflectance with type of cell backing occurs, it is concluded that light is penetrating the cell content completely, and thus is partly lost. In such a case the "true reflectance value of the sample is not obtained." Robinson *et al.* (1952) made further studies on the matter and found that a ¾-in. cell depth was sufficient for the least-opaque juices. To the present authors it seems that a definition of the "true reflectance," requiring independence of the type of cell backing, might be very sound from the theoretical point of view. However, as far as the measurement of the true reflectance of tomato juice is concerned, it is not considered a very important issue and in some cases might even be found impractical. Moreover, it is felt that the color range of raw tomato juice as evaluated from "true" reflectance measurements might differ little, if any, from the color range as determined by the technique described in this paper. Nevertheless, when reflectance measurements are made the conditions of measurement should be made known. In the event that cell size and backing would become an important factor in reflectance measurements of tomato juice, consideration should be given to the relation between reflectance, scattering, and opacity of the juices. Such a relationship has been studied for other materials (Judd, 1952).

To clarify the effect of the color range on the cell depth an auxiliary experiment was performed with the spectrophotometer following the same procedure given above. Six samples of canned tomato juice differing widely in color were each measured in five containers of different depths. The typical results obtained for one of the six samples are given in Table 1. It is evident that even a 10-mm cell depth does not significantly affect the color coordinates of the juice sample as measured in cells of greater depth.

Table 1. Effect of cell depth on chromaticity coordinates x and y and luminous reflectance Y of tomato juice

Cell depth (mm)	x	y	Y
10	0.515	0.357	8.2
25	0.516	0.357	8.3
50	0.514	0.357	8.3
100	0.516	0.356	8.3
150	0.515	0.356	8.3

RESULTS AND DISCUSSION

1. *Effect of holding time and deaeration treatment on the color of raw tomato juice.*

Table 2 summarizes the effects of holding time on the color of untreated raw tomato juice prepared from individual tomatoes of different stages of maturity. It can be seen that the changes in color are a function of time, and the largest changes occur in the juice samples of more advanced maturity. The changes are most pronounced in the Hunter "a" value, indicating a change toward less-saturated and darker colors.

Table 2. Effect of holding time on the color of untreated raw tomato juice prepared from individual tomatoes of different stages of maturity

Maturity	Holding time (min)	Color change[a]			Standard deviation		
		ΔL	Δa	Δb	$\sigma\Delta L$	$\sigma\Delta a$	$\sigma\Delta b$
Light red	30	.6	1.0	.4	.4	.6	.2
	60	.7	1.2	.5	.5	.8	.3
	90	.9	1.5	.6	.6	.8	.4
Medium red	30	.5	.9	.3	.1	.2	.1
	60	.7	1.3	.4	.3	.4	.2
	90	.9	1.7	.5	.3	.6	.2
Dark red	30	.6	1.4	.4	.5	1.3	.3
	60	1.1	2.4	.7	.8	2.0	.6
	90	1.3	3.0	.9	.9	2.5	.6

[a] Each value represents the average obtained from duplicate readings on 4 individual juice samples. The direction of change is toward darker and more saturated colors.

Table 3 summarizes the effects of holding time on the color of raw tomato juice after addition of two drops of n-octyl alcohol. Compared with the color changes given in Table 2, the effects of holding on the color of juice treated with n-octyl alcohol are similar but somewhat less pronounced. The direct effect of the n-octyl alcohol treatment as measured on a few dark-red samples before and immediately after the

addition of n-octyl alcohol produced the following differences in Hunter values:

ΔL	Δa	Δb	$\sigma^{\Delta L}$	$\sigma^{\Delta a}$	$\sigma^{\Delta b}$
.5	.7	.4	.1	.3	.1

The direction of change was toward higher values of L, a, and b with a slight increase in the a/b ratio.

Table 4 summarizes the effects of holding time on the color of raw tomato juice deaerated with vacuum treatment. The results show that vacuum-treated juice was affected only slightly by holding periods up to 90 minutes. The changes occur in the opposite direction as compared to those reported in Tables 2 and 3, indicating a slight change toward more-saturated and lighter color. However, the standard deviations are on the same order of magnitude as the differences in L, a, and b values. The change in color is not considered significant. The direct effect of vacuum treatment as measured on a few dark red samples was as follows:

ΔL	Δa	Δb	$\sigma^{\Delta L}$	$\sigma^{\Delta a}$	$\sigma^{\Delta b}$
1.2	2.6	.8	.3	.2	.1

These differences are three times as large as those obtained for juices treated with n-octyl alcohol. The direction of change, however, is again toward higher L, a, and b values. The increase in the a/b ratio essentially confirms measurements of Yeatman and Sidwell (1958).

Microscopic observations indicated that the n-octyl alcohol removed most of the larger air bubbles, but a great number of smaller bubbles remained. Increasing n-octyl alcohol to 3 or 5 drops per 100 ml juice did not change the results significantly. On the other hand, the vacuum treatment did remove practically all air bubbles after about 15 repetitions of alternate pulling and breaking the vacuum.

Table 3. Changes in Hunter L, a, and b values of juice samples after addition of 2 drops of n-octyl alcohol and subsequent holding at room temperature for 30, 60, and 90 min

Maturity	Holding time (min)	Color change[a]			Standard deviation		
		ΔL	Δa	Δb	$\sigma^{\Delta L}$	$\sigma^{\Delta a}$	$\sigma^{\Delta b}$
Light red	30	.4	.7	.3	.1	.1	.1
	60	.5	.6	.4	.2	.1	.2
	90	.5	.7	.5	.2	.1	.1
Medium red	30	.3	.9	.2	.1	.6	.2
	60	.4	1.1	.5	.4	.9	.2
	90	.5	1.5	.8	.4	.7	.4
Dark red	30	.2	.9	.2	.1	.4	.2
	60	.3	1.2	.5	.2	.7	.2
	90	.4	1.4	.6	.3	.6	.2

[a] Each value represents the average obtained from duplicate readings on 4 different juice samples prepared from 4 individual tomatoes. The direction of change is toward darker and more saturated colors.

Table 4. Effect of holding time on the color of raw tomato juice deaerated with vacuum treatment immediately after pressing

Maturity	Holding time (min)	Color change[a]			Standard deviation		
		ΔL	Δa	Δb	$\sigma^{\Delta L}$	$\sigma^{\Delta a}$	$\sigma^{\Delta b}$
Light red	30	−.1	−.4	−.1	.1	.1	.1
	60	−.1	−.2	−.1	.1	.2	.1
	90	−.2	−.3	−.2	.1	.2	.1
Medium red	30	−.1	−.3	−.1	.1	.1	.1
	60	−.2	−.4	−.2	.2	.2	.1
	90	−.2	−.5	−.1	.1	.2	.1
Dark red	30	−.1	−.1	−.1	.1	.1	.1
	60	−.1	−.2	−.1	.1	.1	.1
	90	−.1	−.2	−.1	.2	.2	.1

[a] Each value represents the average obtained from duplicate readings on 4 juice samples prepared from 4 individual tomatoes.

2. *The color range of raw tomato juice.*

The results obtained in the study of the color range of raw tomato juice as determined by variety and maturity are summarized in Figs. 2 and 3. Fig. 2 shows a portion of the C.I.E. x,y chromaticity diagram into which the chromaticities of 21 samples of raw tomato juice of variety Ferguson are plotted. Underneath those chromaticities an insert is made in the diagram to show the luminous reflectance Y (in per cent) for each of the 21 samples as compared to its x-chromaticity value. The samples marked 1, 2, and 3, shown in open circles and triangles, are respectively representatives of dark-red, medium-red, and light-red juices. These samples respectively represent the higher, the intermediate, and the lower degree of maturity. Plotting their respective a,b values into a score diagram as given by Kramer *et al.* (1959), the samples would obtain the respective color scores of 97, 89, and 78. This means that samples 1 and 2 are within the range of "fancy" grades (86–100), sample 2 being close to the border of "fancy" and "standard," and sample 3 within the range of "standard" grades (77 to 85) but close to the "substandard" range (up to 76).

Two important features of the color range of individual raw tomato juices as determined by maturity may be seen clearly from Fig. 2. The chromaticities seem to follow a somewhat curved line in the diagram, with samples of high degree of maturity at the right-hand side of the curve and samples of lower degree of maturity at the left. Such an order is not surprising when one considers the change in the proportion of green and red pigments present in a tomato

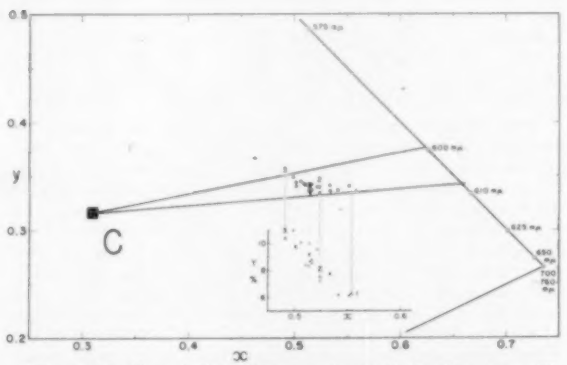

Fig. 2. Portion of the C.I.E. x,y chromaticity diagram, with chromaticities of 21 raw tomato juice samples of variety Ferguson. Insert of an x,Y diagram illustrating the correlation between "redness" and "lightness" of raw tomato juice. Samples 1, 2, and 3, shown by open circles and open triangles, refer to tomato juice of high, intermediate, and low maturity. The solid square "C" is the x,y location of reference illuminant "C." The portion of the spectrum locus covered by the diagram is marked with a few wavelengths given in m_μ.

fruit at various levels of maturity. The other interesting feature in the color gamut of raw tomato juice is the very high correlation of maturity with lightness, as shown in the insert. For the variety Ferguson (Fig. 2), the correlation coefficient was calculated to be .90. With increasing x-values, which may be correlated with an increase in redness, there can be observed a decrease in luminous reflectance Y that may be correlated with a decrease in lightness. As may be expected, there is some scatter in the measurements, but nevertheless the correlation may be considered an important characteristic of the color range of raw tomato juice.

Results with respect to chromaticity coordinates and luminous reflectance were very similar for the other four varieties, listed previously. Because of their close similarity, only the chromaticity loci of these varieties are illustrated in Fig. 3. In each case it was verified that the correlation between x and Y was similar to that shown in Fig. 2 for the Ferguson variety. Therefore this relationship is omitted in Fig. 3.

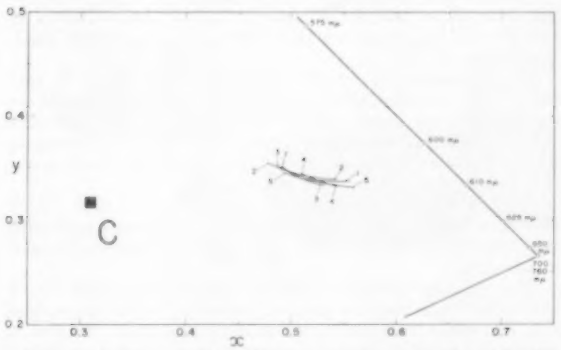

Fig. 3. Portion of the C.I.E. x,y chromaticity diagram with average chromaticity loci of tomato varieties. The solid square "C" is the x,y location of reference illuminant "C." The portion of the spectrum locus covered by the diagram is marked with a few wavelengths given in mμ.

To check on a possible seasonal effect the chromaticity coordinates of the individual juice samples were examined in relation to the dates of picking the fruits. However, no correlation could be detected.

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Effect of Blanching on Texture and Pectin of Canned Cauliflower

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SUMMARY

The degree of esterification of pectic substances in cauliflower is decreased by activation of pectinesterase (PE) during low temperature-long time (LTLT) blanching. As compared with HTST-blanching an LTLT-blanch causes a greater firmness, which is enhanced by addition of Ca-salts and some acid.

The following process is recommended: LTLT-blanching (e.g. for 15 min. at 158° F), cooling, followed by vacuum treatment in a 1.0–1.5% $CaCl_2$-solution. After removal of vacuum cauliflower should be kept in the solution for at least 5 min. and is then washed to remove excess of $CaCl_2$. After packing in cans and adding a brine, containing 1% NaCl, 0.05% citric, and 0.03% ascorbic acid, the product is sterilized by a botulinum cook.

INTRODUCTION

THE ADDITION of Ca salts may be a valuable aid to the improvement of firmness of canned vegetable products (Durocher and Roskis, 1949; Loconti and Kertesz, 1941). This applies particularly to those products in which an important part of the pectic substances have a relatively low degree of esterification and are therefore precipitated by Ca salts (Loconti and Kertesz, 1941).

Tissue characterized by pectic substances with a rather high degree of esterification are firmed to a small extent by the addition of Ca salts. Soluble pectins with a high degree of esterification, such as those from apples, are not flocculated by Ca salts.

According to Deuel *et al.* (1950) soluble pectins can be precipitated by Ca salts in certain cases if the degree of esterification is lower than 50%. Also of influence is the manner of distribution of the esterified groups along the pectic chains. Pectins that have been de-esterified partly by pectinesterase (PE) appear to be more sensitive to Ca salts than pectins de-esterified in an acid or alkaline milieu (Mottern and Hills, 1946). During the action of PE, only adjacent esterified groups are attacked (Mottern and Hills, 1946, Solms and Deuel, 1954), whereas the de-esterification in acid or alkaline milieu occurs at random (Demain and Phaff, 1957; Mottern and Hills, 1946).

The pH of a product during heat treatment as well as the degree of flocculation of the pectins is important to the final firmness. Structural differences in the cell walls of the processed products also play a rôle, though exact information about this factor has not been found (Doesburg, in press).

In some cases the firmness of various canned vegetable products may be improved by treatment with Ca salts or by altering the pH. Such treatments proved inadequate for good firmness in canned cauliflower, however (Hoogzand and Moerman, 1958).

EXPERIMENTAL

Firmness of canned cauliflower. The initial pH of cauliflower is approx. 6.0. Investigations have revealed that cauliflower can be acidified to pH ±5.0 without harming the taste. After blanching (3 min 212° F) the product was brought to this pH by dipping into an acid solution and then retorted 30 minutes at 240° F. It appeared that the decrease in pH did not improve firmness appreciably.

Though blanching in Ca salt solutions somewhat improved the firmness of the finished product, the resulting increase in firmness was still insufficient. As it was thought to be possible that during blanching (3 min 212° F) insufficient calcium would be incorporated, the blanching time was lengthened. Therefore the blanching temperature was lowered, because a prolonged blanch at higher temperatures would cause an undesired loss in firmness.

The effects of some blanching methods were compared: *a)* 15 min in water at 158° F; *b)* 15 min in 2% $CaCl_2$ solution at 158° F; *c)* 3 min in water at 212° F; *d)* 3 min in 2% $CaCl_2$ solution at 212° F.

These experiments revealed that after retorting (30 min 240° F), the firmness of the canned products was improved by the use of a low temperature–long time (LTLT) blanch in a $CaCl_2$ solution. However, it appeared that LTLT blanching in water, without the use of $CaCl_2$, also caused an improvement in firmness after canning and sterilization.

Continued experiments revealed that after a similar LTLT blanch, in contrast with the effect of a high temperature–short time (HTST) blanch, firmness was favorably affected by lowering the pH.

The results of the use of Ca salts and lower pH after HTST and LTLT blanching are given in Table 1. In this experiment only the small thickened flower clusters were used. They were very well mixed, to ensure a uniform product and provide a perfect constant ratio between solid and liquid portions of the can contents.

The following treatments were compared: *a)* blanching in water for 15 min at 158° F or 3 min at 212° F; *b)* similar LTLT or HTST blanching, followed by dipping of the blanched product in 1½% $CaCl_2$ solution under vacuum; *c)* blanching and treatment as performed under *a* and *b*, followed by addition of 0.033% citric acid and 0.033% ascorbic acid to the brine, or no addition of acids.

The weight ratio of product to brine was 3:5. After brining the cans were sealed and retorted 25 minutes at 240° F.

Averages of pH and firmness measurements on the contents of 5 cans per treatment are given in Table 1. Firmness as expressed in kg, was measured with the hardness meter (Doesburg, 1960, Sterling, 1959).

Table 1. Influence of blanching, calcium treatment, and addition of acid on firmness of canned cauliflower

Blanching conditions	$CaCl_2$ treatment	Addition of acid	pH after steriliz.	Hardness after steriliz. (kg)	mg Ca in 100 g total solids
15 min/70° C	−	+	5.28	6.1	463
15 min/70° C	−	−	6.23	1.8	488
15 min/70° C	+	+	5.18	10.1	1146
15 min/70° C	+	−	6.00	4.4	1168
3 min/100° C	−	+	5.40	0.2	457
3 min/100° C	−	−	6.29	0.1	416
3 min/100° C	+	+	5.21	2.6	1121
3 min/100° C	+	−	6.08	0.0	1277

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The figures show that firmness was greatly improved by an LTLT blanch followed by application of Ca salts and acidification of the brine. The combination of both of the latter factors and LTLT blanching produced the greatest degree of firmness.

Effect of blanching on pectic substances and pectin esterase (PE) in cauliflower. The greater effect of Ca salts after LTLT blanching suggests that these blanching conditions lower the degree of esterification of pectic substances.

To investigate whether this hypothesis is correct, pectin analyses were made on cauliflower samples blanched by various methods.

The thickened small flower clusters of some cauliflowers were cut off and mixed; different methods of blanching were applied. To compare the special effect of these blanching methods with the results of a conventional HTST blanch (4 min 212° C), the application of the special blanching conditions to different aliquots was followed by a heat treatment for 4 min at 212° F (Table 2).

After blanching and heat treatment the cauliflower parts were ground 3 min in a Waring blender with an equal weight of acetone. The suspension obtained was filtered over a Buchner funnel. The solid parts were treated with acetone to the weight of the first suspension, and filtered again. This treatment was repeated once more, after which the cauliflower was dehydrated at 104° F.

The dehydrated product was weighed and then ground to a powder in a ball mill.

Pectin in the cauliflower-powder samples was extracted with water. The weight of the powder-water mixture was always twice that of fresh cauliflower.

In a few cases the total quantity of pectin was determined by acidifying the powder suspension with HCl to pH 3.3 and then boiling for 30 min (Table 2, experiment A). The water suspension was then diluted with water to the original weight and filtered over a Buchner funnel.

The pectin content and its degree of esterification in the filtrate were determined by a titrimetrical method (Doesburg, 1957).

In other cases (Table 2, experiments B and C) the soluble pectin was extracted first by soaking the powders for 3 hours at 86° F in water as mentioned above, and subsequent mixing of the suspensions for 3 min in a Waring blender. Then the suspensions were filtered over a Buchner funnel. In the filtrate the amount of soluble pectins was determined by the titrimetrical method.

Water-insoluble pectin in the residue on the filter was extracted with 0.5% ammonium oxalate solution. The pH was adjusted to 3.3 with conc. HCl, and the suspension boiled for 30 min. The liberated pectins were determined as mentioned above.

Originally, we tried to isolate the pectic substances in three fractions: *1)* the fraction soluble in water; *2)* the fraction soluble in NH_4-oxalate solution; *3)* the fraction to be isolated by boiling at pH 3.3. However, it appeared that after extraction of soluble pectin, the second fraction—to be isolated by 3 hours' extraction in a ½% NH_4-oxalate solution at 122° F—was negligible. For these reasons the two last fractions were extracted together, as described above.

To investigate whether the decrease in esterification of pectin after LTLT blanching could be attributed to the activity of PE, a preparation of this enzyme from cauliflower was tried with the technique described by Edwards and Joslyn (1952).

Of the extracts thus obtained, 4 ml was added to 20 ml of an apple pectin solution adjusted to pH 6 with a borate-acetate buffer. Immediately after mixing, boiling tubes with these mixtures were placed in a water bath at different temperatures for 15 min followed at once by a 4-min treatment in boiling water. Then the tubes were rapidly cooled. The results of the analyses of the pectin solutions thus treated are given in Table 3. Data are given on the influence of PE solution inactivated by previous heating to 100° C. PE preparations of cauliflowers from different sources gave almost identical results.

Table 3. Action of PE extract from cauliflower on an apple pectin solution

Heat treatment of the pectin solution	PE extract		Boiled PE extract	
	Esterification (%)	Pectin content (mg/100 g)	Esterification (%)	Pectin content (mg/100 g)
4 min 212° F	68.9	94.5	70.2	94.1
15 min 176° F plus 4 min 212° F	61.3	91.2	.....	.....
15 min 158° F plus 4 min 212° F	59.5	89.0	68.5	93.3
15 min 140° F plus 4 min 212° F	53.3	72.5	.....	.....
15 min 122° F plus 4 min 212° F	48.6	59.5	.....	.....

DISCUSSION

It has been shown that improved firmness in canned cauliflower by the LTLT blanch is related to the decrease in degree of esterification of the pectic substances.

It is well known (Doesburg, in press; Doesburg and Grevers, 1960) that the degree of esterification is decreased by heating in low acid or neutral milieu. Comparison of the influences of various blanching temperatures on the degree of esterification (Table 2) is hampered by the fact the times applied have been different for various temperatures. It is unlikely that the lower degree of esterification resulting from an LTLT blanch is caused by the longer saponification period in low-acid milieu. The data in Table 3 show that in the heat-inactivated PE extracts the degree of esterification is affected to only a small degree by heating 15 min at 158° F. The LTLT blanching process gave a decrease in esterification greater in the insoluble pectin fraction than in the soluble pectin (Table 2); this fact indicates that de-esterification was not caused by the heat treatment as such.

As stated in a preliminary report (Doesburg, 1959; Hoogzand and Moerman, 1958), the lower degree of

Table 2. Influence of various blanching conditions on pectic substances in cauliflower

Method of blanching	Experiment A		Experiment B				Experiment C			
	Total quantity of pectic substances		Water-soluble pectin		Insoluble pectin		Water-soluble pectin		Insoluble pectin	
	Content (%)	Degree of esterification (%)	Content (%)	Degree of esterification (%)	Content (%)	Degree of esterification (%)	Content (%)	Degree of esterification (%)	Content (%)	Degree of esterification (%)
4 min 212° F	0.20	70.3	0.08	57.7	0.22	55.8	0.12	63.3	0.23	50.6
4 min 194° F plus 4 min 212° F	0.23	69.0	.....	.....	.....	.....	0.14	64.1	0.20	48.3
4 min 176° F plus 4 min 212° F	0.25	66.3	0.12	56.3	0.21	54.8	.....	.....	.....	.....
15 min 158° F plus 4 min 212° F	0.19	63.2	0.13	52.7	0.22	43.6	0.10	59.5	0.25	39.3
15 min 158° F, 1 hour cooling, 4 min 212° F	0.21	62.7	.....	.....	.....	.....	.....	.....	.....	.....
30 min 140° F plus 4 min 212° F	.....	.....	0.08	52.8	0.24	48.4	.....	.....	.....	.....

esterification after LTLT blanch is caused by the activation of PE such as that always found in cauliflower. If Ca salts are used in the blanching water, the greater firmness from LTLT blanching than from HTST blanching, is not brought about by a greater Ca incorporation in the LTLT-blanched product (Table 1).

Since PE has been found in many plant species (Deuel and Stutz, 1956; Kertesz, 1951), it was to be expected that LTLT-blanching could produce the same effect in various products. This view may be confirmed by Van Buren *et al.* (1960), who found similar relations between blanching and firmness in snap beans. They postulated that firmness is related to enzyme action. It is very likely that there has also been a PE activation in snap beans.

The data in Table 3 reveal that the optimum temperature of the isolated PE was lower than 158° F. Table 2 shows that in situ the activity of PE is lower at 140° F than at 158° F.

The pectinase activity, which was found in the PE preparations, was not demonstrated in the blanched cauliflower (Table 2).

The action of all PE preparations from cauliflower on apple pectin solutions also caused a decrease of the amount of pectin (Table 3). In cauliflower the partial de-esterification during blanching was not accompanied by a decrease of pectin content (Table 2). However, during blanching some limited depolymerization of pectin chains in cauliflower may have occurred, which is not shown by pectin analysis (Doesburg, 1957).

According to Henglein and Krohn (1949) the swelling power of dead vegetable tissue might depend on the degree of esterification and the degree of polymerization of the pectic substances; thus, swelling increases with a higher polymerization or degree of esterification.

The cauliflower powder, as prepared for pectin analysis, showed different degrees of swelling if the cauliflower was blanched in different ways. These differences in swelling power were visible during filtration in the Buchner funnel, and can be demonstrated by suspending the powders in water.

Fig. 1 shows the difference in swelling ability of the two samples in water (5% solution, w/v) prepared from cauliflower blanched 4 min at 212° F, or blanched 15 min at 158° F, followed by 4 min at 212° F. The less evident swelling of the LTLT-blanched sample can be attributed to the lower degree of esterification of the pectin substances.

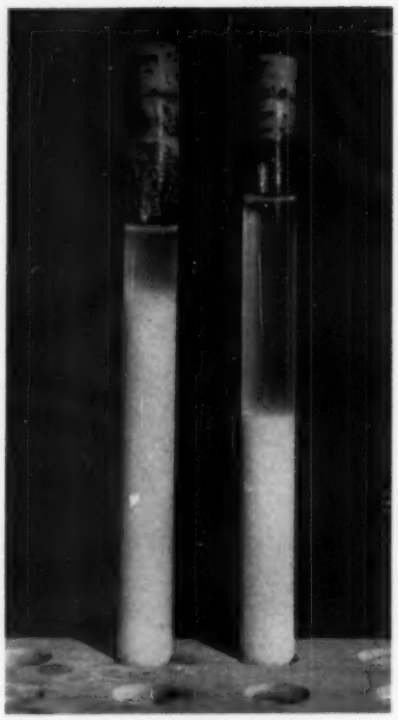

Fig. 1. Swelling of powder, made of cauliflower after application of different blanching conditions (1 g powder in 20 ml water). Right: Blanching 15 min. 158° F, followed by heat treatment 4 min. 212° F. Left: Blanching 4 min. 212° F.

CONCLUSIONS

Firmness of canned cauliflower is better from a 15-min blanch at 158° F than from a blanching for 4 min at 212° F. The use of Ca salts and addition of a small quantity of acid to the brine increase the firmness. The following process is recommended for a canned product with good firmness and natural color: *1)* low temperature–long time (LTLT) blanching (e.g., 15 min at 158° F); *2)* vacuum treatment of the blanched and cooled cauliflower in a 1.0–1.5% $CaCl_2$ solution; after removal of vacuum, the cauliflower should be kept in the solution for at least 5 minutes and then washed to remove the excess $CaCl_2$; *3)* packing the product in cans and adding a brine containing 1% NaCl, 0.05% citric acid, and 0.03% ascorbic acid; *4)* sterilization of the canned product by application of a botulinum cook.

The favorable effect of LTLT blanching can be attributed to the activation of pectinesterase (PE) in cauliflower, which lowers the degree of esterification of the pectic substances. Partial de-esterification of apple pectins by activity of PE preparations from cauliflower was always accompanied by a decrease of the pectin amount. In cauliflower, however, partial de-esterification during blanching was not connected with a decrease of pectin amount.

Acknowledgment

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Stability of Carotenoids of Dehydrated Sweet Corn on Storage

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(Manuscript received September, 1960)

SUMMARY

Golden Cross Bantam sweet corn contains several carotenoid pigments. Two major components were found in the endosperm, and were tentatively identified as lutein and zeaxanthin. Lutein predominated in the embryo. On storage of dehydrated samples, whether sulfited or not, there was no demonstrable change in the carotenoid content at any stage of maturity tested. Nevertheless, there may be a critical stage in the maturation process that must be passed before carotenoid stability can be assured.

THERE HAVE BEEN several studies on the carotenoid pigments of sweet corn (e.g. Petzold and Quackenbush, 1960; White *et al.*, 1942a, b) and a series of papers dealing with sweet corn dehydration (e.g. Gardner *et al.*, 1955; Hayes *et al.*, 1956). There have also been suggestions that the carotenoids of dehydrated sweet corn tend to bleach on storage, and according to von Loesecke (1955) the bleaching is believed to be due to selective absorption of sulfur dioxide by the kernels.

We have tested the effects of maturity, processing variables (blanching and sulfiting), and storage (in air, under nitrogen, and in vacuum, at 0, 37, and 47°C for periods up to 70 days). There was no demonstrable change in carotenoid content as a result of processing or storage, and a brief note recording these findings seems warranted. As noted by Booth (1957), material low in carotenoid is often difficult to extract and "the seeds of *Zea mays* are especially intractable." The problem of proper extraction is therefore critical.

EXPERIMENTAL

Material. The corn for all studies was Golden Cross Bantam. Also, a large-scale test was conducted at the J. R. Simplot plant at Caldwell, Idaho. The corn came from nearby fields and was graded. Quantities needed to obtain 40 lb cut corn were: immature, middle A grade or higher, 240–260 ears; mid-mature, bottom A, 180 ears; mature, middle to top B, 150 ears.

Extraction of Coloring Matter. The non-uniform distribution of coloring matter as observed under the microscope led us to treat embryo and endosperm separately in preliminary tests. Heating greatly facilitated extraction of pigment from the embryo, but had less effect on ease of extraction from the endosperm.

Fresh samples of whole kernel, usually *ca* 10 g, were blended

with distilled water for 5 minutes, acetone was added to make a concentration of 90%, the slurry was blended and filtered, and the residue was re-hydrated and re-extracted four times. This brought yields of extracted pigment to *ca* 80% of the level attainable if the kernel had previously been given a 5-minute blanch in boiling water. Unlike extracts from processed material, the coloring matter could not be completely transferred to petroleum ether but was readily transferable to diethyl ether. Extracts were then saponified, washed, and dried with Na_2SO_4.

Dehydrated samples of whole-kernel corn were powdered in a Wiley mill, 5-g samples were weighed and rehydrated overnight with 10 ml water at 5°C, 90 ml acetone were then added, and the mixture was allowed to stand 24 hours at 5°. It was then filtered and the residue washed with acetone. The white residue thus obtained yielded no further pigment on re-hydration and re-extraction.

Dehydration Test at Caldwell, Idaho. For each maturity, 40-lb samples were given blanches in steam (208°F at that altitude) of zero, 3.5, and 10 minutes. Sulfite concentrations of zero, *ca* 2500, 5,000, and 10,000 ppm yielded respective final concentrations in the dry product of *ca* 2–300 (due to small quantities of sulfur in the fuel), 1000, 2000, and 4500 ppm.

The corn was dehydrated on stainless-steel trays in a Proctor and Schwartz Pilot Forced-Air Dryer.

RESULTS

Embryo and Endosperm. Total carotenoid was estimated at 11.7 μg per g for the embryo, and 5.5 μg for endosperm. Calculations were based on optical densities at the maximum between 440 and 450 mμ, and an arbitrary approximation of 250 L per g cm for the coefficient for the carotenoid mixture. Since the weight ratio of endosperm to embryo was about 4 : 1, the distribution of pigment between endosperm and embryo is on the order of 2 : 1.

By chromatography of endosperm extracts on MgO-SiO_2 columns, 8 components were separated; two were tentatively identified (by absorption spectra, adsorption behavior, and partition coefficients) as zeaxanthin and lutein. They were present in approximately equal amounts and together accounted for 70% of absorption at 450 mμ. Three components were separated from embryo extracts. The zeaxanthin was missing; the lutein comprised 65% of the total, on the same basis.

Whole kernel. In a test with a saponified extract in the Craig Countercurrent Distribution Apparatus, between Skelly-Solve B, methanol and water (1640, 900, 9 ml), the diol-polyol fraction showed a maximum concentration, after 100 transfers, at tube numbers $N_{10\text{-}11}$, and constituted *ca* 74% of the total, monols (cryptoxanthin and zeinoxanthin) N_{55}, *ca* 20%; hydrocarbons (including β-carotene, Z-carotene, phytofluene) N_{90}, *ca* 6%.

The Caldwell Tests. Results were completely at variance with the hypothesis that the SO_2 would destroy the carotenoid. The over-all average for 75 analyses, at Berkeley, regardless of treatment or maturity, was 6.75 ppm $\pm$ 11%. The averages for immature, mid-mature, and mature were respectively 6.34 $\pm$ 12.3%, 7.05 $\pm$ 8.5%, and 6.91 $\pm$ 9.4%.

Of 38 analyses at Caldwell, omitting four that were clearly inconsistent, the over-all average was 6.10 ppm $\pm$ 16%. The lower value may be attributed to lack of a small mill to reduce particle size to ensure complete extraction.

DISCUSSION

We have observed that carotenoids in solution are bleached in contact with SO_2, but in the tissue in which they are laid down they appear to be relatively stable. Huelson (1954) records minor losses of carotenoids in dehydrated sweet corn in storage up to 4.5 years, whether samples were sulfited or not. Deterioration was greatest in immature samples, however.

Acknowledgment

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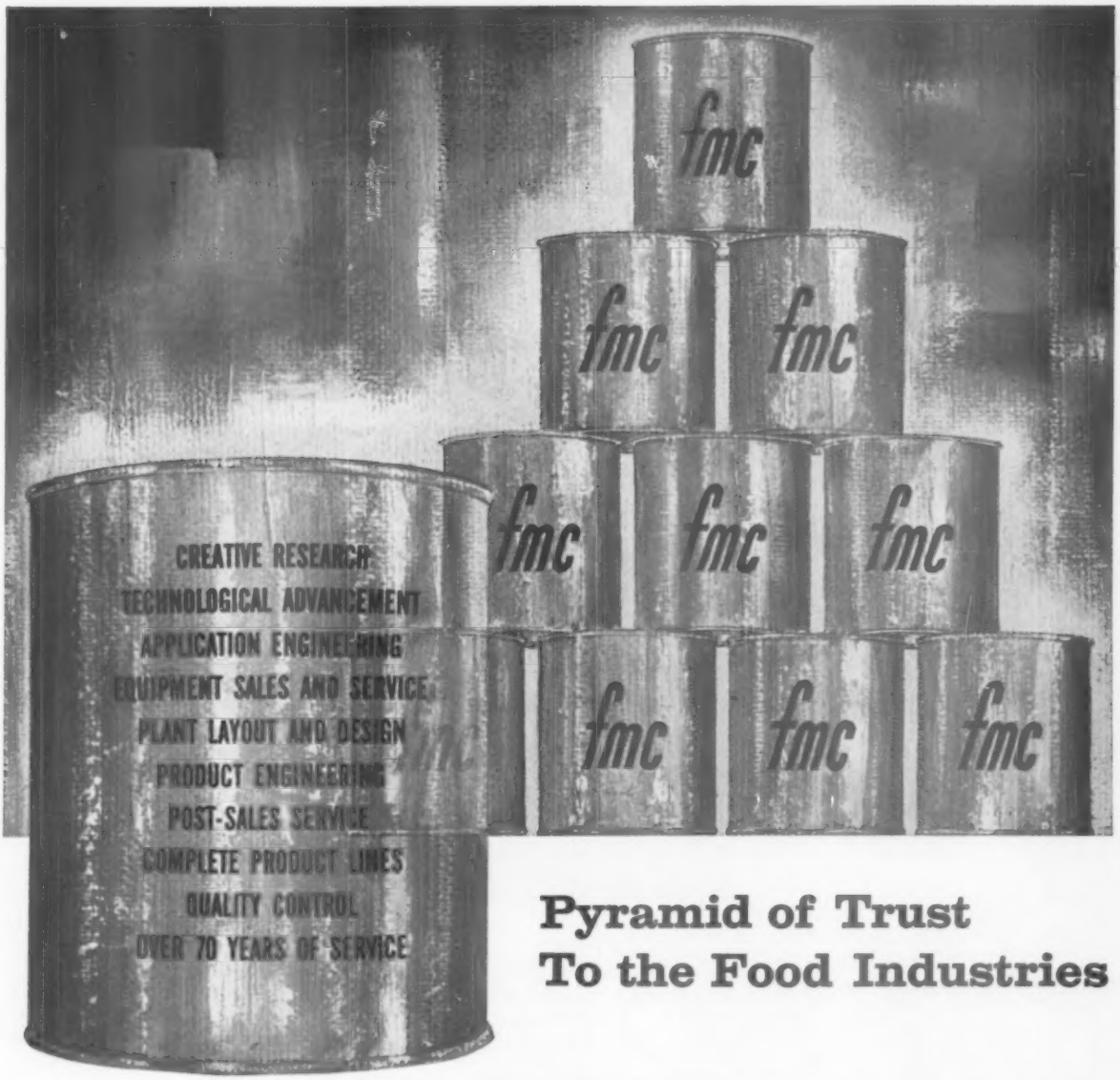

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Volatile Reducing Substances as a Criterion of Quality of Canned Apricots*

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(Manuscript received September 10, 1960)

SUMMARY

The influence of ripeness level and 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) spray on the chemical composition and the volatile reducing substances (VRS) of canned apricots was studied. The aroma of canned apricots improved as the fruit ripened. The VRS content in canned apricots paralleled the aroma rating of the canned product. At the same ripeness level, the control samples were slightly lower in VRS than the 2,4,5-T–sprayed samples. The acetyl-methyl-carbinol and methyl alcohol contents of the canned apricots increased as the fresh fruit ripened, especially in the samples of the low pressure range that were sprayed with 2,4,5-T. The importance of ripeness level to aroma and VRS of the canned product is discussed.

THE QUALITY of canned apricots is related to the ripeness of the fresh fruit at the time of canning (Hoos *et al.*, 1956). Pressure tests and Brix/acid ratios are used as criteria for determining ripeness. Spraying apricot trees at the pit-hardening stage with 2,4,5-trichlorophenoxyacetic acid (2,4,5-T) is considered horticulturally advantageous because of the resulting increase in fruit size, earlier maturation, and prevention of preharvest fruit drop (Crane, 1954; Crane and Brooks, 1952).

Allen (1932), Atkinson and Strachan (1940), and Strachan *et al.* (1951) reported on the relation of maturity to the chemical constituents of fresh apricots. Kramer and Smith (1947) used electrophotometric methods for measuring ripeness and color in canned peaches and apricots. The chemistry of fruit and vegetable flavors has been reviewed by Kirchner (1949), but information on the volatile constituents of apricots is very limited. Farber (1949) reported on the chemical evaluation of odor intensity by the permanganate titration method, but gave no information on volatile constituents in canned apricots.

Herein is reported the influence of ripeness level and 2,4,5-T spray on both the chemical composition and the volatile reducing substances (VRS) of canned apricots.

MATERIALS AND METHODS

Cultural treatments. Each of three Blenheim apricot trees at the University orchard in Winters, California, was sprayed at the pit-hardening stage with 6 gal. of an aqueous solution of 2,4,5-T at a concentration of 100 ppm (0.79 ml of tri-ethyl amine salt of 2,4,5-T per gallon of water). Three trees that received no 2,4,5-T spray were used as a control. All received ⅔ lb of nitrogen and were irrigated once. About 450 lb of fruit was harvested from the sprayed trees. The fruit was sorted into four ripeness levels by skin color.

Canning. The apricots were washed in tap water, sorted for ripeness by skin color, and halved and pitted in a Filice and Perrelli apricot pitter. Those in the over-ripe stage, being too soft for mechanical pitting, were halved and pitted by hand. Nineteen and a half ounces of fruit and 10.5 oz. of 40% sucrose syrup were added to each No. 2½ can. The cans were sealed under 15 in. of vacuum, cooked 19 minutes at 212°F in a rotary cooker, and cooled rapidly in water for 7 minutes. The canned product was stored at room temperature for one month before chemical and organoleptic evaluations were made.

Pressure test, titratable acidity, soluble solids, and pH. These values were determined in the manner described previously (Leonard *et al.*, 1953; Magness and Taylor, 1925).

Aroma. A panel of ten persons judged the aroma of the canned product with three replications. Each time, four samples were presented to the panel, and the judges were requested to score them for aroma as follows: excellent, 9–10; good, 7–8; fair, 5–6; poor, 3–4; and very poor, 1–2. The results were analyzed for variance as described by Snedecor (1946).

Volatile reducing substances. VRS content of the canned apricots was determined by the potassium permanganate oxidation method (Luh *et al.*, 1955) modified as follows: The contents of three cans of chilled apricots were mixed for 30 seconds in a Waring blender. Two hundred and fifty grams of the sample was steam distilled until 200 ml of distillate was collected. The distillate was diluted with distilled water to 250 ml. Duplicate samples of 75-ml portions were treated for 30 minutes in glass-stoppered Erlenmeyer flasks with 20 ml of 0.1*N* $KMnO_4$ in 1*N* NaOH. Ten ml of 5*N* sulfuric acid and 10 ml of 30% potassium iodide were then added to the flask. The iodine liberated was titrated with 0.1*N* sodium thiosulfate using a 1% soluble starch solution as the indicator. Results were reported as microequivalents of $KMnO_4$ per 100 g of sample. A blank was run using 250 ml of distilled water instead of the sample.

Methyl Alcohol. A 2-ml portion of the above distillate was used to determine the methyl alcohol content by the permanganate-chromotropic acid method described by Boos (1948).

Total carbonyl compounds. A 1-ml portion of the distillate was analyzed for total carbonyl compounds. This was done by measuring the color developed from 2,4-dinitrophenylhydrazine in alkaline solution, as explained by Lappin and Clark (1951).

Voges-Proskauer reaction. Twenty-ml portions of the distillate were analyzed for their acetyl-methyl-carbinol content by the Voges-Proskauer reaction method (Barritt, 1936) described by Byer (1954).

RESULTS

Chemical analyses and aroma scores of canned apricots. The effect of ripeness level and 2,4,5-T spray on the chemical composition and aroma of canned apricots was investigated. Data presented in Table 1 indicate that samples canned at higher pressure tests were lower in Brix reading and pH than samples canned at lower pressure tests. The titratable acidity of the fruit decreased as the ripening process proceeded. Both the control and the sprayed series showed the same trend.

The control samples were smaller (50.0–55.4 g av weight per apricot) than the sprayed ones (64.6–67.7 g). This confirms reports (Crane, 1954; Crane and Brooks, 1952) that 2,4,5-T spray increases the fruit size.

* This study was supported by the United States Public Health Service, National Institute of Health, Grant RG-5800.

Table 1. Effect of ripeness level and 2,4,5-T spray on chemical analyses and aroma of canned Blenheim apricots (Winters, California).

		Fresh apricots		Canned apricots				
Sample	Treatment	Average pressure test of fresh apricots (lb; 5/16-in. plunger)	Average weight of individual fruits (g)	Brix at 20°C	Titratable acidity as % citric acid	pH	Brix/acid	Aroma score[a]
1	Control	3.1	52.9	21.5	1.06	3.60	20.3	4.4
2	Control	2.2	52.0	22.6	1.00	3.65	22.6	4.1
3	Control	1.0	55.4	23.8	0.96	3.75	24.8	6.7
4	Control	0.3	50.0	24.4	0.88	3.76	27.7	8.3
5	2,4,5-T spray	4.1	66.9	20.8	0.92	3.75	22.6	3.7
6	2,4,5-T spray	2.0	64.7	21.8	0.87	3.84	25.0	4.4
7	2,4,5-T spray	1.3	64.6	22.1	0.82	3.86	27.0	6.3
8	2,4,5-T spray	0.5	67.7	23.2	0.77	3.99	30.1	7.6

[a] Higher score represents better apricot aroma; LSD = 1.5 at 95% probability level.

Brix/acid ratio is related to the ripeness levels of the fresh fruit. In the control series, the ratio increased from 20.3 (Sample 1) to 27.7 (Sample 4) as the average pressure test of the fresh fruit decreased from 3.1 to 0.3 lb. A similar increase in Brix/acid ratio with ripening was observed in the 2,4,5-T–sprayed series.

Aroma is one of the attractive characteristics of canned apricots. Table 1 shows that aroma of the canned apricots improved as the fruit ripened. Fruit canned at a soft-ripe stage was scored better in aroma than fruit canned at a firmer stage. It is evident that the ripeness level of fresh fruit is related to the aroma of the canned product.

Volatile reducing substances. Fig. 1 shows total VRS content of Blenheim apricots from the Winters area, canned at different pressure levels. It is expressed as μeq of $KMnO_4$ per 100 g of sample in which the fruit and syrup were combined prior to analysis. As the apricots ripened and the pressure level decreased, the total VRS content increased. The increase was more rapid when the pressure test dropped below 2 lb. In the 2,4,5-T–sprayed series in which the pressure test at canning decreased from 4.1 to 0.5 lb, the total VRS content in the canned product increased from 125 to 575 μeq of $KMnO_4$ per 100 g of sample. At the same firmness level, the control samples were lower in VRS than were the sprayed samples. It appears that 2,4,5-T spray caused the production of a higher VRS content in the fruit. Fig. 2 indicates that the acetyl-methyl-carbinol content of canned apricots increased rapidly as the fresh fruit ripened, especially in the samples of the low-pressure range that were sprayed with 2,4,5-T.

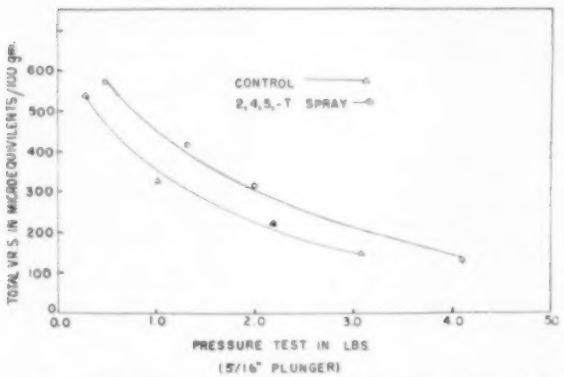

Fig. 1. Volatile reducing substances in Blenheim apricots canned at different ripeness levels.

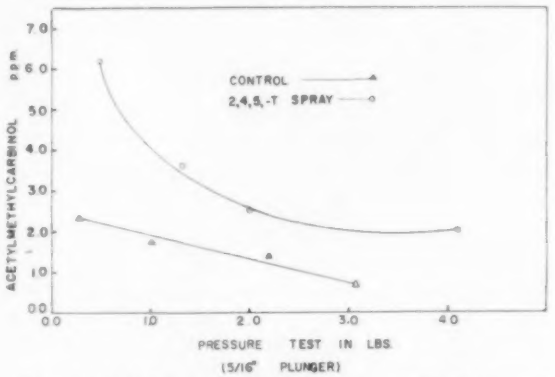

Fig. 2. Acetyl-methyl-carbinol content of Blenheim apricots canned at different ripeness levels.

The methyl alcohol content also increased with ripening of the fresh apricots (Fig. 3). The increase was more rapid when the pressure test was at one pound or less. At the same ripeness level, the samples sprayed with 2,4,5-T were higher in methyl alcohol content than were the control samples.

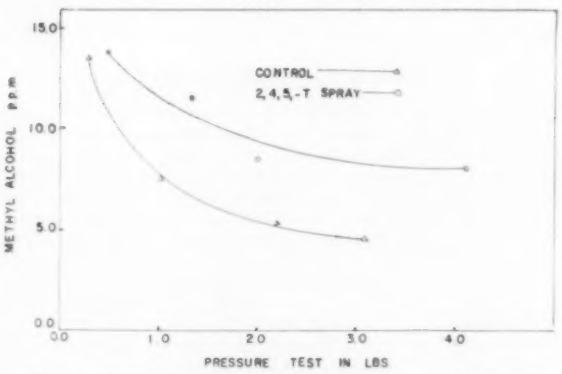

Fig. 3. Methyl alcohol content of apricots canned at different ripeness levels.

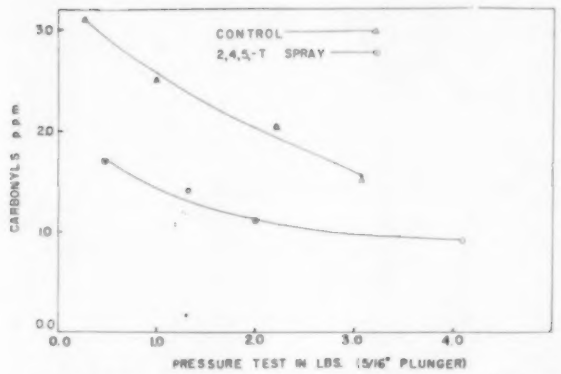

Fig. 4. Carbonyl compounds in apricots canned at different ripeness levels.

The amount of carbonyl compounds increased as the pressure reading of the fresh fruit decreased (Fig. 4). At comparable pressure tests, however, the control samples contained a higher amount of carbonyl compounds than did the 2,4,5-T–sprayed samples.

It is important to point out that the volatile compounds did not increase at the same rate as the firmness decreased. Apparently, various metabolic processes were proceeding at different rates as the fruits ripened on the tree. Some of the VRS was accumulated as end products of such processes.

DISCUSSION

The determination of VRS may help in evaluating the odor intensity of food (Farber, 1949). This was found to be applicable to canned apricots. Samples with higher VRS contents were rated better in aroma. Fruits canned below 2 lb pressure test were better in aroma than fruits canned at a firmer stage. As the fruits ripened, however, the canned product tended to have a ragged appearance due to tissue breakdown. When apricots are not of uniform ripeness, it is desirable to select those at optimum ripeness level for canning, and the over-ripe, soft fruit for nectar, concentrate, preserves, or other products that do not require whole fruit tissue for a good appearance.

The amount of volatile compounds in apricots was shown to be influenced by the 2,4,5-T spray, indicating that biological processes in the fruit might be influenced by application of the growth regulator. The quality of the canned products is determined largely by the ripeness level at canning, although the 2,4,5-T spray caused a slight increase in VRS content.

The increase in methyl alcohol content during ripening may be explained partly by the hydrolysis of the methyl ester group in the pectin by the enzyme pectinesterase. This was shown to be true in Bartlett pears (Luh *et al.*, 1955). However, the amount of methyl alcohol found in canned ripe apricots was only 15 ppm, whereas in a ripe pear sample it was 96 ppm (Luh *et al.*, 1955). This indicates a great difference in the properties of these two fruits.

Only a few of the many possible VRS of Blenheim apricots are reported on in this paper. However, the increase in total VRS correlated well with the decrease in fruit firmness and improvement in aroma score of the canned product. Once the range of total VRS for normal samples has been established and correlated with organoleptic data, it may be possible to evaluate objectively the effect of horticultural practices, growing area, post-harvest handling, and ripening on the quality of the canned product. The data presented here, though of a rather cursory nature, serve to illustrate that poor quality in canned apricots can be evaluated objectively in terms of chemical composition.

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Lipid Oxidation in Pre-Cooked Beef Preserved by Refrigeration, Freezing, and Irradiation[a]

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SUMMARY

Oxidation of lipids in the lean tissue of roast beef slices preserved by refrigeration, freezing, or irradiation was followed by the thiobarbituric acid test and organoleptic evaluation. Oxidized products accumulate very rapidly in the refrigerator. Frozen samples maintain a somewhat lower oxidation over long storage periods. Lipid oxidation is not an important factor in irradiated beef stored at room temperature. Antioxidant combinations of ascorbate and polyphosphate, used either as dips or as cover solutions, eliminate lipid oxidation and greatly improve the odor of refrigerated and frozen beef, but do not benefit irradiated beef.

PREVIOUS WORK from this laboratory has shown that heat treatment of uncured meats initiates in the tissue lipids oxidative changes that result in rapid development of stale or rancid off odors (Tims and Watts, 1958). The lipid fractions chiefly involved are not the triglycerides, but rather proteolipids and phospholipids (Younathan and Watts, 1960). The reaction is believed to be catalyzed by the ferric heme pigments of cooked meats but not by cured meat pigments (Younathan and Watts, 1959). Inhibition of oxidation with appropriate antioxidants greatly improves the odor ratings of both refrigerated and irradiated cooked ground pork (Tarladgis *et al.*, 1959).

All work so far reported on this type of oxidative deterioration in cooked meats was done on ground meat, usually cooked in closed containers but with headspace oxygen. Using ground meat was simpler because it allowed uniform mixing and sampling of large batches and because antioxidants could be incorporated thoroughly.

Further work has demonstrated that such lipid oxidation is even more of a problem in large cuts of meat cooked whole and later sliced and exposed to air. The present paper explores the importance of this reaction in roast beef slices preserved by refrigeration, freezing, or irradiation. Antioxidant dips, cover solutions, and variations in oxygen availability are investigated as methods of control.

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METHODS

Preparation of meats. Used exclusively was the Longissimus dorsi (rib eye) muscle from beef ribs. The fresh meats were purchased at local retail markets. For any one experiment, the meat used was from one muscle only, or from paired muscles of a single animal.

In an oven set at 325°F the muscles were heated uncovered to an internal temperature of 160°F (71°C). This required about 2 hours. They were cooled rapidly by wrapping in aluminum foil and immersing in ice water, and were then sliced with a mechanical slicer to thicknesses ranging from 4 to 7 mm in different experiments. The slices were treated, packaged, and stored as described in each experiment. Room temperature was about 25°C, refrigeration temperatures varied from 1 to 4°C, and the freezer was set at −26°C.

Tissue lipid oxidation and odor. Rancidity of tissue lipids was measured at various storage intervals with the 2-thiobarbituric acid test (Tarladgis *et al.*, 1960). Duplicate 10-g samples of meat freed of visible fat were used for each sampling period. The results were expressed as "TBA number," i.e., mg of malonaldehyde per 1000 g meat. Odor evaluations on a scale of 1–6 (very strong to no off odor) were made by trained panels as previously described (Tarladgis *et al.*, 1959).

EXPERIMENTAL RESULTS

EXPERIMENT 1. *Odor deterioration in exposed slices of roast beef.* Slices were removed from a fresh roast at intervals starting 4 hours before the judges were assembled to rate the samples. The slices were exposed to the air in covered containers at room temperature. The last slice, freshly cut just before the judges assembled, was designated zero exposure. The results (Table 1) clearly show the rapid development of rancid off odor in exposed surfaces of roast beef.

Table 1. Expt. 1. Effect of exposure to air on sensory scores in roast beef.

Exposure (hr)	Sensory score[a]
0	5.7 (a)
1	4.6 (b)
2½	4.1 (c)
4	2.9 (d)

[a] a>b, c, d (differences highly significant).

EXPERIMENT 2. *Effect of antioxidant dips on roast beef slices stored in the refrigerator and the freezer.* In previously reported experiments on ground meat the addition of 0.5% sodium tripolyphosphate and 0.1% ascorbate to the raw meat was sufficient to give complete protection from oxidative changes during the refrigerated life of the product (Tims and Watts, 1958) and during extended storage of irradiated meats (Tarladgis *et al.*, 1959). Primal cuts obviously cannot be protected in this way. This experiment was

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undertaken to determine if dipping slices of the cooked meat in an aqueous antioxidant solution would protect them. Since the antioxidant was to be applied as a brief dip to the surface of the meat only, the concentrations of polyphosphate and ascorbate were respectively increased to 1.0 and 0.27% of the solution.

Slices were dipped immediately in the antioxidant solution (or in distilled water as control) for 30 seconds and drained for 10 seconds. All slices were folded into 307 x 113 enamel cans, about 150 g of meat per can. The cans were sealed at atmospheric pressure and stored either in the refrigerator or the freezer.

At various storage intervals, corresponding samples were prepared for analysis by blending the entire contents of the can with an equal weight of water. Aliquots of the resulting slurry representing 10 g of meat were used for the TBA test. Organoleptic tests and pH measurements were also made on the slurry. The respective pH values of the cooked control and antioxidant-treated samples were 5.6 and 6.1. Other results are in Table 2.

Table 2. Expt. 2. Effect of antioxidant dips on roast beef slices stored in the refrigerator and the freezer.[a]

Storage temperature	Days in storage	No antioxidant (control)		Dipped in 1% $Na_6P_2O_{10}$ + .27% Na-ascorbate	
		TBA no.	Odor score	TBA no.	Odor score
1°C	1	11.5	....	0.3	....
	4	15.2	2.5	0.4	5.6
	7	19.5	2.6	0.3	5.3
	11	21.1	1.9	0.3	4.7
	18	21.2	....	0.3	....
—26°C	11	3.5	2.7	0.2	5.4
	31	2.3	2.4	0.2	5.5
	45	3.2	....	0.2	....
	81	4.9	2.2	0.2	5.9
	164	4.1	3.5	0.2	5.8

[a] At each storage period, the odor of the antioxidant-dipped meat was rated against its corresponding control. In all cases, the antioxidant-dipped rated higher than the control, and the differences were highly significant.

With no antioxidant protection, the refrigerated samples showed a progressive increase in TBA number to a maximum of 21, which is higher than the maximum values (7–10) previously obtained from ground meat after cooking and refrigerator storage. In frozen samples without antioxidants, the TBA numbers fluctuated irregularly between 2 and 5. Odor scores of the frozen samples were not very different from those of the refrigerated samples having much higher TBA numbers, but it must be remembered that the judges compared each control sample only with the antioxidant-treated sample at the same storage period. At only one storage period (11 days) were both refrigerated and frozen rated at the same time, and here the odor scores were higher for frozen than for refrigerated samples.

The antioxidant treatment gave striking protection to both refrigerated and frozen meats. The TBA numbers remained well below the organoleptically detectable threshold value, about 0.5–1.0 (Tarladgis *et al.*, 1960). The original pleasant odor of the freshly cooked meat was maintained throughout the storage period.

EXPERIMENT 3. *Effect of antioxidant dips on irradiated roast beef slices.* This experiment was designed to determine whether the antioxidant dips that stopped lipid oxidation in refrigerated and frozen beef (Expt. 2) would also benefit irradiated roast beef slices. For this purpose, samples were prepared as described in the last experiment. The pH of the water-dipped samples was 6.1. The antioxidant dip gave an increase to 6.65.

The sealed cans were packed in dry ice and shipped by air express to Argonne National Laboratory. Half the cans received 4.8 megarads of gamma irradiation at ambient temperature. The remaining cans were not irradiated, but were otherwise handled in the same manner as the irradiated ones. After irradiation, all cans were returned in dry ice. The unirradiated control cans and half of the irradiated cans were then stored in the freezer. The remaining irradiated cans were stored at room temperature.

TBA values at various storage periods are presented in Fig. 1. The unirradiated frozen samples

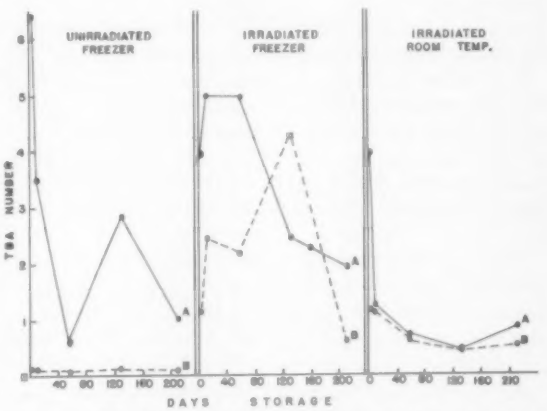

Fig. 1. Effect of antioxidant dips on lipid oxidation in unirradiated and irradiated roast beef slices. Solid line, control (water dipped); broken line, dipped in antioxidant (1% sodium tripolyphosphate + 0.27% sodium ascorbate).

behaved as in the previous experiment, except that the initial TBA values were higher in the absence of antioxidant, probably because of the additional thawing at the Argonne. The lower TBA numbers of the irradiated samples stored at room temperature, as compared to those in the freezer, were associated with development of pink color at the higher storage temperature, whereas the frozen samples remained gray or greenish gray.

Odor ratings were made on all samples at four storage intervals ranging from 0 to 210 days of storage. As in the previous experiment, unirradiated samples dipped in antioxidant were much fresher in odor than the controls. Average odor score (all storage periods) was 5.8 for the antioxidant-dipped samples and 4.2 for the controls. The difference is highly significant.

Odor scores for the irradiated samples, in contrast, were not improved by antioxidant dips or by freezer versus room-temperature storage. The average score was 3.6 (range 3.0–4.4), with no significant difference between any two treatments at any storage period. All had a strong irradiation odor.

EXPERIMENT 4. *Effect of cover solutions.* Since it has been well established that oxygen is involved in the rancidification of unsaturated fatty acids and in the irradiation-induced oxidative damages, covering the sample with liquid to reduce contact with oxygen might be expected to have some favorable effect. An experiment was planned to investigate 1) the use of covering solutions, and 2) the increase of antioxidant concentration.

Roast beef slices were prepared as in the last experiment. Before they were folded into the cans, some slices were dipped either in a solution of 2% sodium tripolyphosphate and 0.5% sodium ascorbate or in distilled water. Others were covered either with the above solution or with distilled water, 100 ml per can. On their return from the Argonne, unirradiated samples were stored in the freezer, and irradiated samples at both room and freezer temperatures. The samples were analyzed at different storage periods. The results are in Table 3. The meat slices instead of the slurry were used for odor comparisons.

The cover solutions, both with and without antioxidants, gave lower TBA values than the corresponding dip techniques in all freezer-stored samples. With the irradiated samples, as in the previous experiment, there was little difference in any of the samples after room-temperature storage; all TBA numbers were low.

Data on odor ratings are incomplete because of difficulties in assembling sufficient judges. The cover solutions improved the odor scores of the frozen irradiated samples, but not of those stored at room temperature.

All unirradiated samples were the typical gray-brown of cooked meat. In all covered irradiated samples, the color appeared as bright pink at all times after irradiation. As long as the meat was still in the solution, opening the can did not cause fading of the color within a few hours of observation. Fading was rapid once the meat was exposed directly to air.

EXPERIMENT 5. *Evacuation and oxygen packing.* Comparable slices of roast beef were irradiated and stored under rather extreme conditions, i.e., in evacuated containers versus containers flushed with oxygen. The slices were prepared and dipped as in Experiment 2. Each was placed in a 5 x 6-in. flexible package (polyethylene-coated polyester film, laminated to 0.001 aluminum foil, laminated to 18 lb plasticized sulfate paper, manufactured by the Minnesota Mining and Manufacturing Company). The packages were partially sealed, evacuated or flushed with oxygen through an inserted glass tube, and then sealed completely. Variations in the amount of oxygen from package to package are to be expected with this procedure.

The irradiation source in this experiment was a new 3-mev Van de Graaff electron accelerator on this campus. The electrons were not used as such but were transformed to X-rays by impinging on a gold target. Eight packages could be irradiated at one time in a fairly uniform field at a dose rate of about 2.1 megarads per hour. The total dose given the samples was 3.4 megarads. (It was not intended to irradiate below the currently accepted sterilization dose of 4.8 megarads, but at the time the experiment was performed the dose rate had not been accurately measured.) Unirradiated samples were stored in the freezer, and irradiated samples at room temperature. Data are presented in Table 4.

Packaging in oxygen resulted in greater lipid oxidation in both frozen and irradiated samples. Irregular variations in the TBA numbers at different

Table 4. Exp. 5. Effect of variations in oxygen.[a]

Radiation treatment	Antioxidant treatment	Storage time	Evacuated		Oxygen added	
			TBA no.	Odor score	TBA no.	Odor score
None (stored in freezer)	Control	Upon return	0.1		13.5	
		12 days	4.4		5.9	
		28 days	0.7	4.5	11.6	2.8
	Antioxidant dip	Upon return	0.2		0.2	
		12 days	0.1		0.1	
		28 days	0.1	5.9	0.1	5.6
3.4 megarads (stored at room temperature)	Control	Upon return	1.0		11.3	
		12 days	0.8		8.6	
		47 days	0.4	5.0	10.4	2.4
	Antioxidant dip	Upon return	0.3	4.0	1.4	4.3
		12 days	0.3		2.4	
		47 days	0.6	4.4	2.9	2.4

[a] Statistically significant differences in odor scores: Unirradiated: antioxidant treated > controls (highly significant); evacuated control > oxygenated control (significant). Irradiated: At 47 days evacuated > oxygenated (highly significant).

Table 3. Expt. 4. Effect of cover solutions on lipid oxidation and odor.

Sample treatment	Storage time	TBA number				Odor scores			
		Control	Antiox. dipped	Water cover	Antiox. cover	Control (1)[a]	Antiox. dipped (2)[a]	Water cover (3)[a]	Antiox. cover (4)[a]
Unirradiated stored in freezer	Upon returning	3.4	0.1	1.0	0.1	....	....	....	....
	53 days	3.7	0.4	1.0	0.1	4.0	....	....	5.7
Irradiated stored in freezer	Upon returning	2.7	0.9	2.1	0.4	3.8	....	....	4.5
	185 days	2.2	0.6	0.6	0.3	3.3	3.8	4.5	5.2
stored at room temp.	53 days	0.5	0.9	0.4	0.5	3.1	....	....	3.1
	185 days	0.4	0.2	0.2	....[b]	....	....	....	....

[a] Code numbers. Statistical differences for odor scores at 185 days expressed by code numbers are as follows: 4>1, 2, 3, highly significant; 3>1, 2, significant; 2>1, not significant.
[b] Defective seal.

storage periods undoubtedly resulted from the non-uniformity of packaging. Other work has shown that when cooked fish was stored in sealed containers, TBA numbers quickly reached limiting values determined by the amount of oxygen present, and remained at these values indefinitely (Florida State Univ., unpublished). The use of antioxidant dips completely protected the frozen samples, even under these extreme conditions, but were much less effective than evacuation for the irradiated samples.

Exclusion of oxygen in the irradiated samples resulted in a bright-pink, whereas oxygen caused destruction of pigment, with a resulting gray color. These color differences became progressively more evident during storage.

DISCUSSION

It is apparent that the oxidation of tissue lipids proceeds at a very rapid rate when roast beef is sliced and exposed to the air in the refrigerator or at room temperature. TBA numbers are regularly 2- to 3-fold higher for such exposed slices than for ground meat heated to the same internal temperature, although the reason why is not clear. Similar experiments on roast pork, lamb, and veal also resulted in very high TBA numbers (20 to 30) and stale odors in the lean meat after several days' exposure in the refrigerator. Oxidation is very much slower in the freezer than in the refrigerator. It is possible that unavoidable exposure to higher temperatures during preparation for freezing and especially during thawing may account for at least part of the oxidation observed, although precautions were taken to blend and analyze frozen samples without delays. Other work has shown that TBA values shoot up very rapidly when precooked meats are removed from the freezer and stored in the refrigerator (Florida State Univ., unpublished).

Control of this reaction, with remarkable improvement in odor of the preserved meat, can be achieved with the antioxidant combination of tripolyphosphate and sodium ascorbate, used either as a dip or as a cover solution. The antioxidant concentrations used in this work were chosen arbitrarily. Experiments in progress indicate that, for the normal refrigerated or frozen storage life, the polyphosphates alone, at a dip concentration of 0.5%, may be sufficient to give complete protection. A range of concentrations of these and other antioxidants needs to be investigated.

The antioxidant mixture used is more alkaline than the normal pH range of meat, and the treated meats were about 0.5 pH unit higher than the corresponding controls. However, the change in pH is not an important factor in controlling lipid oxidation. Variations in pH over a much wider range produced no significant differences in the rate of lipid oxidation in the absence of antioxidants (Florida State Univ., unpublished).

The implications of these experiments for the improvement of meat items in precooked frozen dinners are obvious. It is also possible that a greatly extended line of refrigerated precooked meat products, appropriately sliced and packaged, could be made available in retail stores. Most of the table-ready meats currently displayed contain curing salts, and consequently are not subject to the rapid flavor deterioration of uncured cooked meats (Younathan and Watts, 1959). The rather simple antioxidant dip treatment described here would make possible uncured products of good flavor, without the necessity of smoking, covering with strongly flavored sauces, or excluding oxygen from the package. The ease of protection could also improve leftover meats, prepared meat sandwiches, etc., in the home.

On the other hand, beef preserved by sterilizing doses of ionizing radiations apparently cannot be improved by the antioxidant treatments. Lipid oxidation products did not contribute a significant fraction of the irradiation-induced odor complex except under conditions of abnormally high oxygen pressure. On storage at room temperature, the rate of lipid oxidation decreased to insignificant values, concomitant with the development of pink reduced heme pigments which are believed to lack the catalytic activity of the ferric cooked meat pigments (Tarladgis *et al.*, 1959). Under conditions where lipid oxidation was measurable in irradiated beef, antioxidant dips were ineffective in retarding oxidation.

It is quite possible that much lower doses of irradiation, in the "pasteurizing range" of 0.1–1.0 megarads, coupled with suitable antioxidant treatments, could result in well flavored precooked beef products with a greatly extended refrigerated life. It is also possible that antioxidant dips or cover solutions would improve radiation-sterilized cuts of pork or chicken, which have much less irradiation odor than beef.

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Notes and Letters

The Formation of Pheophorbides During Brine Preservation of Cucumbers

Willstatter and Hocheder (1907) observed a change of the chlorophylls to pheophytins by the action of dilute acids, resulting in replacement of the complexly bound magnesium in the chlorophyll molecule with hydrogen and a coincident change in pigment color from green to brown. Green vegetables usually undergo a similar color change from green to brown during processing and storage. Several investigators (Blair and Ayers, 1943; Campbell, 1937; Dietrich *et al.*, 1959; Dutton *et al*, 1943; Fishbach and Newburger, 1943; Legault *et al.*, 1950; Mackinney and Weast, 1940; Sweeney and Martin, 1958) have reported the conversion of chlorophyll to pheophytin by processing procedures involved in the preservation of green vegetables by canning, freezing, and dehydration.

Studies in progress on the chlorophyll changes that take place in green cucumbers during brine preservation show that the brown chlorophyll derivative that forms during brining is largely composed of pheophorbides *a* and *b* rather than pheophytins *a* and *b*. The pheophorbides are pheophytins from which the phytyl group has been removed. Pheophorbides may be formed by the action of the enzyme chlorophyllase (Weast and Mackinney, 1940; Willstatter and Stoll, 1928a).

The pheophorbides may be identified or separated from the pheophytins by extraction of ether solutions of the pigments with 0.01 molar potassium hydroxide solutions. The pheophorbides are soluble in the basic aqueous solution (Willstatter and Stoll, 1928c).

In this study the pheophorbides were extracted from ether solutions of the chlorophyll derivatives. The quantitative relationship of pheophorbides and pheophytins formed during the brining of cucumbers is shown in Table 1, representing some typical observations which have been made.

Table 1. Absorbance of pigments in diethyl ether at 666.5 mμ

Before extraction with 0.01*M* KOH. Pheophytins and pheophorbides	After extraction with 0.01*M* KOH. Pheophytins	Difference representing pheophorbides
0.800	0.146	0.654
0.730	0.135	0.595
0.697	0.125	0.572
0.722	0.133	0.589
0.767	0.144	0.623

The identity of pheophorbides *a* and *b* was verified by determining the acid numbers of the extracted pigments according to the procedure described by Willstatter and Stoll (1928b). Considerable pheophorbide *a* was extracted from an ethereal solution of the pigments by 15% HCl, giving a bluish-green solution. Extraction of the same ethereal solution with 22% HCl gave a yellowish-green aqueous solution of pheophorbide *b*.

The removal of the phytyl group from the chlorophyll molecule or its conversion product during the brining process was enzymatically induced, as was shown by blanching studies. Cucumber tissue that had been adequately blanched before brining yielded chlorophyll conversion products consisting principally of the pheophytins rather than the pheophorbides.

Studies are in progress to establish the path of conversion of the chlorophylls to the pheophorbides during the brining process, and the rate of such change.

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ANNUAL MEETING NEWS

Eight Field Trips Planned

Eight field trips are scheduled for Thursday, May 11, to some of the numerous food industry firms and educational institutions in the New York area. Two of the featured tours will last all day, while the other six will be divided into three morning and three afternoon trips.

The "North" all-day tour, from 8 A.M. to 5 P.M., includes a visit to the General Foods Research Center at Tarrytown. Each of the anticipated one hundred participants will be the guest of General Foods for lunch. The afternoon phase will entail a trip to Fairlawn, N. J., where National Biscuit Company will be host.

The "South" all-day tour, with a maximum attendance of eighty individuals, encompasses several areas of endeavor of interest to the food technologist. Stops will be made at Rutgers—The State University and the Walker Gordon plant, located at New Brunswick and Plainsboro, N. J. respectively. Third stop will be at Firmenich, Inc. where participants will be luncheon guests of the International Flavor house. Prior to returning to the Hotel, a visit will be made to the Princeton campus. This tour will also be from 8 A.M. to 5 P.M.

Six individual food industry firms will be visited on each of the other field trips. Scheduled for the morning are Hoffmann-La Roche, with maximum attendance of thirty-five. Visitors will be shown through the Nutley, N. J. plant and then participate in a luncheon—courtesy, Hoffmann-La Roche.

The second and third morning trips are each expected to be filled to the capacity of 45 individuals. Rahway, N. J., location of Merck & Co., host for a tour and luncheon, is the site of one of the trips, while International Flavors and Fragrances, Inc., Union Beach, N. J., will host the other. The IFF group will be guests of that organization for a luncheon at Buttonwood Manor in South Jersey.

Both the Roche and Merck Tours are expected to be completed by 1:15 P.M., while the IFF Tour will return at 2:00 P.M.

Final tours of the Convention, each accommodating 45 persons, will commence at 1:30 P.M. First afternoon tour will be a visit to Thomas J. Lipton, Inc.,

SPACE AGE FLAVORS...

This is the day of jet-flight and dehydrated potatoes; atomic energy and frozen bread; super-sonic speed and "quicker-than-instant" mixes for everything from soups to desserts. This is the threshold of the space age and the era of convenience foods. Flavors for these modern foods must be as dynamic as the technology of their formulation . . . as carefully designed as the science that developed them. Yet, these space-age foods must still retain that home cooked taste. Only a company that keeps thoroughly abreast of the newest developments in food can give you this kind of "built for the product" flavor. Fries & Fries provides you with space-age flavors with that country kitchen taste! Let our laboratories show you what dynamic flavor research means.

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• Flavorex Jamaica Style Rum is a flavor concentrate of natural and synthetic esters produced in our own plant and blended by our masters of the art. Run a batch test. You'll discover that every fragrant drop adds mellow tropical sunshine to your product. Write for free sample, or place a trial order today.

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Hoboken, N. J. Second and third will be to Food and Drug Research Laboratories, Inc., Maspeth, L. I., N. Y., and Anheuser-Busch, Newark, N. J.

Simple arithmetic will reveal that a combined total of 440 individuals can be accommodated on all field trips. Members will receive advance field-trip request forms in the Annual Meeting invitation envelope, and an early return of the request will aid in ensuring that you may participate in the trip most suited to your desires.

BOOK REVIEW

ADVANCES IN PROTEIN CHEMISTRY, Vol. 14, C. B. Anfinsen, Jr., M. L. Anson, Kenneth Bailey and John T. Edsall, Ed., Academic Press, New York, 1959.

This volume contains seven review articles that should be valuable to fundamental researches in food science. The three articles that appear to have the most probable value to food researchers are "Some Factors in the Interpretation of Protein Denaturation," by W. Kauzmann, "Zone Electrophoresis in Starch Gels and Its Application to Studies of Serum Protein," By O. Smithies, and "The Sulphur Chemistry of Proteins," by B. Cecil and J. R. McPhee. Kauzmann's comprehensive article emphasizes the changes in fundamental structures of the native protein that occur as the molecule becomes unfolded during denaturation. The article is primarily divided into two sections, one on methods of observing changes in the configurations of polypeptide chains, and one on the forces responsible for maintaining the native configuration of proteins. Denaturation, of course, is pertinent to all food processes that involve thermal treatments. The article by Smithies is an excellent presentation of the new tool of starch-gel electrophoresis, the principal developments of which he has been responsible for. The article contains experimental procedures for employing starch-gel electrophoresis as well as a discussion of its application and consideration of some of the theoretical aspects. Starch-gel electrophoresis has made possible separations of previously unresolvable complex mixtures, and should have many uses in food research. The article by Cecil and McPhee should have particular interest because it presents a critical study of the methods of determining disulfide and sulfhydryl groups, important to the physical and flavor properties of foods. The other four reviews are: "The Specificity of Protein Biosynthesis," by M. Vaughan and D. Steinberg, "Structural Aspects of Tobacco Mosaic Virus," by H. Fraenkel-Conrat and L. K. Ramachandran, "The Serum Proteins of the Fetus and Young of Some Mammals," by R. A. Kekwick, and "Biological Properties of Poly-amino Acids," by M. Sela and E. Katchalski.

ROBERT E. FEENEY

CHEMICALS AND THE FOOD INDUSTRY, Robert M. Ikeda and Donald G. Crosby, University of California Division of Agricultural Sciences, Agricultural Experiment Station—Extension Service, Manual 26, June, 1960.

This 127-page manual meets a real need for furnishing a well-organized survey of the current potential use of chemicals in producing, processing, and marketing foods.

The bulk of the manual is devoted to presenting the major technical problems of the food industry. Discussion is by type of food product, and many examples are given to show how chemicals are being or might be used to solve each problem.

Another important section of the booklet lists the additives used by the food industry, tabulated in major categories according to function. The large number and wide variety of substances included here bear testimony to the great importance of chemicals to this industry.

The authors make no attempt to discuss the laws regulating food additives. However, some of the first official lists are reproduced, giving the status of certain chemicals under the Federal Food, Drug and Cosmetic Act, the Federal Meat Inspection Act, and the Federal Poultry Products Inspection Act. Obviously, current regulations must be consulted for up-to-date information.

An excellent foreword has been contributed by Dr. E. M. Mrak, Chancellor, and Dr. G. F. Stewart, Head, Department of Food Science and Technology, University of California, Davis.

This survey is well-documented through 1957, but only a few later references are given. Consequently several new developments are not covered. Nevertheless, the manual will be invaluable to all readers seeking reliable information on the subject of chemicals in foods. Copies of the manual may be obtained for $1.00 each from Agricultural Publications, 207 University Hall, 2200 University Avenue, Berkeley 4, California. HOWARD C. SPENCER

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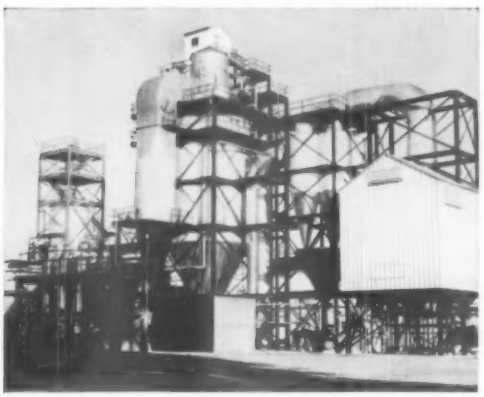

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REGIONAL NEWS
CHICAGO

New Dehydration Techniques Discussed

A panel of four persons concerned with experimental and developmental work in food dehydration discussed new techniques in this field at the January meeting of the Chicago Section.

Dr. Virgil C. Wodicka, Director of the QMF&CI, moderator and panel member for the evening, described briefly two new techniques in dehydrating liquids and slurries. In Europe, particularly in Italy and Switzerland, a new type of installation is in use for spray drying. A tower some 260 feet high is used, with an up-draft of air at 70°F. It is able to dry a wide variety of liquid products, with a "quite reasonable" quality, Dr. Wodicka said.

A technique for dehydrating liquids with a high sugar level has been developed by the Eastern Regional USDA laboratory. The liquid is first evaporated in a conventional film evaporator and the essences collected. Concentration is continued to a point where the solution at a high specific gravity, is a liquid only at high temperatures. The essences are reincorporated into the concentrate and the liquid is then allowed to crystallize on a chilled roller. The sheet breaks off and can be ground into powder. Although there have been no commercial applications, apple juice and grape juice have been dried successfully using this technique. Dr. Roderick Eskew has been in charge of this investigation.

Most of the really new techniques for drying large particles of food have been made in the field of freeze-drying. According to David Rest, food engineering consultant in Chicago, food engineers can now design a system to meet any need.

Storage and transportation of these products give rise to other problems such as oxidative deterioration. One preventive of this is good packaging. Materials for Army purposes have been tested, but no single plastic or combination will suit Army needs, according to Frank J. Rubinate, chief of the development of standards for the QMF&CIAF. The best packaging discovered so far is a three-ply pouch of vinyl film, aluminum foil and mylar. The pouch is protected in an equally strong carton. The result, up to now, is a package so strong that even piano wire will not open it without fail. DENNIS MURPHY

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PHILADELPHIA

Training Course Sponsored by Philadelphia Section

Basic information on food processing practices and methods current in Philadelphia food industries was reviewed for sanitarian staff in a five-day training seminar presented in mid-October by the Philadelphia division of environmental health.

The seminar was designed to provide specific food technology information for a realistic and practical application of sanitation requirements established January, 1960, for food processing plants in Philadelphia.

Assisting Morris A. Shiffman, chief of the milk and food section, in the planning was **Fred Jacobson,** Philadelphia Section President.

Some of the topics and the speakers: "Baking Processes for Bread and Other Yeast Raised Products," Walter Jackson, Penn Fruit Company; "Baking Processes for Cakes and Biscuits," Chris Hansen, American Stores Company; "Macaroni and Macaroni Products," **Murray Cooper,** Insect Control and Research, Inc.; "Candy and Confectionery Manufacturing," **John Vassos,** S. F. Whitman & Son, Inc.; "Meat and Poultry Products," **J. Sam Lafferty,** Oscar Mayer & Company, and "Frozen Food and Cold Storage," David Kronick, Philadelphia Department of Public Health.

Other participants included **Walter Obold,** Drexel Institute of Technology; **Stanley Segall,** Kwik Kafe Coffee Processors; **John Crimmins,** Lummis & Company; **E. Everett Meschter,** American Stores; Robert E. Wing, Tasty Baking Company; Herbert Ayres, Federal Food & Drug Administration, and Delmar Myers, Pennsylvania Bureau of Foods & Chemistry.

FUTURE MEETINGS FOR FOOD TECHNOLOGISTS

1961	
March 20-23	Fifteenth National Conference on Handling Perishable Agricultural Commodities, Horticulture Dept., Purdue University, Lafayette, Indiana
March 26–30	National Association of Refrig. Warehouses, Mark Hopkins Hotel, San Francisco
March 27–31	4th Symposium on Temperature—Its Measurement and Control in Science and Industry, Veteran's Memorial Hall and Deshler-Hilton Hotel, Columbus, Ohio
March 28	Seventeenth Annual Quality Control Clinic, Rochester Society for Quality Control, University of Rochester, Rochester, New York
April 4-6	**Fifteenth Annual Meeting of the Research and Development Associates, Food and Container Institute, Hotel John Marshall, Richmond, Virginia**
April 6–8	Institute of Food Distributors of America, Bal Harbour and Americana Hotels, Miami Beach
April 9–12	U. S. Wholesale Grocers Association, Bal Harbour and Americana Hotels, Miami Beach
April 11-12	Food Engineering Conference, Michigan State University, East Lansing, Michigan. (For information write to Continuing Education Service, Michigan State University, East Lansing)
April 14–18	National Fisheries Institute, Shoreham Hotel, Washington, D. C.
April 15	American Seafood Distributors Association, Shoreham Hotel, Washington, D. C.
April 15	National Shrimp Breeders Association, Shoreham Hotel, Washington, D. C.
April 27–29	Southern and Southwestern Frozen Food Associations, Roosevelt Hotel, New Orleans, Louisiana
May 7-11	**Twenty-first Annual Meeting of the Institute of Food Technologists, Hotel Statler, New York, New York**
May 7–10	Super Market Institute, McCormick Place, Chicago, Illinois
May 15-18	Fourth International Congress on Canned Foods, Berlin, Germany (for information address M. Henri Cheftel, 71, Avenue Edouard Vaillant, Billancourt (Seine), France.
May 17–21	British National Association of Wholesale Distributors of Frozen Foods, Yorks and Old Swan Hotels, Harrogate
May 24–26	Canadian Institute of Food Technology, Queen Elizabeth Hotel, Montreal
May 30	Nutrition Society of Canada, Ontario Agricultural College, Guelph, Ontario
June 13–16	3rd Biennial International Gas Chromatography Symposium, Kellogg Center, Michigan State University
June 15–17	Shrimp Association of the Americas, Mexico City
June 18–23	Association of Food & Drug Officials of the United States, Mayflower Hotel, Washington, D. C.
June 27–30	American Home Economics Association, Auditorium, Cleveland, Ohio
July 10–14	National Food Distributors Association, New York City
Sept. 15–19	American Meat Institute, Palmer House, Chicago, Illinois
Sept. 19–22	Australian Conference on Food Technology, C.S.I.R.O., Homebush, Australia
Sept. 19–27	International Conference on Fish in Nutrition, Washington, D. C.
Sept. 28–30	American Hotel Association, Washington, D. C.
Oct. 7–12	Baking Industry Exposition, Convention Hall, Atlantic City
Oct. 15–18	National Association of Food Chains, Palmer House, Chicago, Illinois
Oct. 18–20	National Packaging Forum of Packaging Institute, Biltmore Hotel, New York City
Oct. 24–27	American Dietetic Association, Jefferson Hotel, St. Louis
Nov. 5–8	National Frozen Food Association, Bal Harbour and Americana Hotels, Miami Beach
Nov. 6–10	National Hotel Exposition, Coliseum, New York City
Nov. 12–15	Air Conditioning and Refrigeration Institute, Homestead Hotel, Hot Springs
Nov. 15	British National Association of Wholesale Distributors of Frozen Foods—Annual Lunch, Trocadero, London
1962	
June 10–14	**Twenty-second Annual Meeting of the Institute of Food Technologists, Fontainebleau Hotel, Eden Roc Hotel, Miami Beach, Florida**
1963	
May 19–23	**Twenty-third Annual Meeting of the Institute of Food Technologists, Pantlind Hotel, Civic Auditorium, Grand Rapids, Michigan**

An open invitation is extended to readers of FOOD TECHNOLOGY to send in to the Editorial Office, P. O. Box 164, Davis, California, notices of annual or national meetings of interest to food technologists.

Literature

BOOKS

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Report on Agricultural Chemicals and Recommendations for Public Policy, Dec. 1960, State of California Governor Edmund G. Brown's Special Committee on Public Policy Regarding Agricultural Chemicals.

Foods for Shelter Storage, A Literature Review for the Office of Civil and Defense Mobilization, 1960, J. G. Woodroof and Olga K. Lebedeff, Georgia Experiment Station, University of Georgia, Experiment, Georgia.

Food Preparation and Processing Equipment Catalog, 1960. Food Machinery and Chemical Corporation, Canning Machinery Division, San Jose, California, Hoopeston, Illinois.

IN MEMORIAM

Dr. William Findlay Geddes, 1958 Nicholas Appert Medallist, passed away on January 7th in Mexico City. During his long teaching career at the University of Minnesota he became endeared to hundreds of students, in addition to creating an international sphere of respect for his research in cereal chemistry. Dr. Geddes served as editor of the journal *Cereal Chemistry*, and is credited with more than two hundred technical papers in his field.

His many affiliations included the IFT, American Chemical Society, American Association for the Advancement of Science, National Research Council committees, the Advisory Board for Quartermaster Research and Development, Sigma Xi, and past president of the American Association of Cereal Chemists.

Honors bestowed on Dr. Geddes include the Coronation Medal of King George VI (1930), the Thomas Burr Osborne Medal of the A.A.C.C. (1950), and the Nicholas Appert Award.

Industry News

A new brominated citrus seed oil, GRAVITOL "C," will be available April 1st from Sterol Derivatives, Inc., 3626 Medford Street, Los Angeles 63, California. According to the manufacturer, it has been fully tested and proven stable in beverage bases, bottled drinks and/or syrups when sunlight-exposed for 21 days, and oven-stored for 45 days at 120°F. In addition, no physical or organoleptic changes were observed when stored in sunlight or at accelerated temperatures. Sterol Derivatives believes that this may prove to be a significant contribution to beverage base manufacturers preferring a citrus-source weighting oil for labeling and product, meeting current or future requirements of the Food Additive Amendment.

Miles Chemical Company, a division of Miles Laboratories, Inc., Elkhart, Ind., has developed a new cellulase enzyme, TAKAMINE Cellulase 4000, to hydrolyze and thus depolymerize cellulosic materials. Current research indicates that the new Miles enzyme may have these applications: brewing—to increase body of finished beers; citrus juices—as a clarifying agent and to increase juice yields; essential oils—to aid in extraction of essential oils and other natural flavors by breaking down plant cell walls; septic systems and drain cleaner—destroy paper and other wastes.

Dr. L. A. Underkofler, director of enzymology at Miles Chemical, noted these research findings have been confirmed:

1. Solutions of the enzyme at its natural pH of 5.0 are indefinitely stable at room temperatures up to 100°F or even higher.

2. The enzyme shows excellent cellulase activity over a broad pH range. Maximum activity is shown at pH 3.0 to 3.5, with good activity at higher or lower acidities.

3. Under relatively acid conditions, pH range 2.2–3.0, from 80 to 95 per cent of the original activity is retained when solutions of the enzyme are held at 100°F for an hour. At pH 3.0 and above no loss of activity occurs under these conditions.

4. The enzyme exhibits unusual thermal stability. At its natural pH of 5.0 a solution of the enzyme retains 88 per cent of its original activity after four hours at 140°F and 43 per cent of its original activity after four hours at 158°F. At pH 3.0 (one per cent acetic acid), 44 per cent of its original activity is retained after four

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Complete Flavors for Prepared Toppings

Supplementary Flavors for Instant Chocolate Drinks

Complete Flavors for Prepared Cake Mixes and Prepared Icings

Yes, the vast number of products that can be benefited by our complete or supplementary flavors is a surprise even to our research department where solving one problem inevitably leads to solving another.

All the way from the many different prepared mixes to instant chocolate drinks, the findings of our food technicians have shown many makers the way to more delicious, distinctive flavors that are individualized for their own product.

We would welcome any questions from Food Chemists about flavor or a visit to our laboratories.

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VANILLA LABORATORIES, Inc.
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1. Eliminate wrong cook schedules.
2. Guard against "by passing" retorts.
3. Warn of any failure in retort processing.
4. Keep retort baskets in sequence.
5. Provide low-cost permanent records for cooking plants.

The cost of Cook-Chex protection is negligible . . . less than one cent for 75 cases of No. 300 cans.

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hours at 125° F, 38 per cent at 135°F and 32 per cent at 145°F. At pH 2.5 (one per cent citric acid), 38 per cent of the original activity is retained after four hours at 125°F.

TAKAMINE Cellulase 4000 is commercially available in unlimited quantities.

Vico 400, a natural fermentation flavor, is said to impart a true yeast-leavened flavor and aroma in chemically leavened baked products without time-consuming dough fermentation. It is especially effective in combination with the blander types of chemical leavening agents, such as glucono-delta-lactone; calcium acid phosphate; cream of tartar and sodium aluminum sulfate, according to the manufacturer. Vico 400 is a highly stable, dry flavoring in powder form, alone or in prepared mixes and frozen dough, said to be easy to handle and ready for use at any time. For sample, technical information and prices, write to: Vico Products Company, 415 West Scott Street, Chicago 10, Illinois.

Paul-Lewis Laboratories, Inc., has announced the introduction of a blue cheese flavor claimed to be totally unique, which is being marketed under the trade name of TRU-BLU. It is said to be the precise quantitative counterpart of the significant flavor volatiles present in the natural product, and is available in plastic and liquid form. Research on the flavor volatiles of blue cheese and use of gas chromatography to follow the development of the volatile flavor components in blue cheese during ripening, aided in bringing about the component identification. TRU-BLU is the combination of those flavor volatiles and as such is an imitation flavor in the form of an actually compounded duplicate of the significant flavor volatiles found in blue cheese. For complete information, write to Paul-Lewis Laboratories, Inc., 4215 North Post Washington Avenue, Milwaukee 12, Wisconsin.

"The following, written by David Kritchevsky, of Philadelphia, was recently published in the *New England Journal of Medicine* . . .

Cholesterol is poisonous
So never, never eat it.
Sugar, too, may murder you
There is no way to beat it.
And fatty food may do you in;
Be certain to avoid it—
Some food was rich in vitamins
But processing destroyed it.
So let your life be ordered
By each documented fact
And die of malnutrition
But with arteries intact."

EXPANSION NEWS

The Anco Specialty Company, Amherst, Ohio, recently acquired manufacturing rights, patents and accounts of the vegetable machinery division of The American Specialty Company, also of Amherst. Since 1930, The American Specialty Company has been manufacturing special vegetable washing equipment for canners, packers, freezers, growers and pre-packers. Anco, metal fabricating specialists, will expand the present line to include such added items as hoppers, bins, chutes, work tables, conveying equipment, exhaust systems and ventilating systems.

Corn Products Company announces that grinding and sales operations have commenced at its newest South American plant, in Uruguay. The plant is equipped to produce edible corn starch, laundry starch and other consumer items, as well as such products for industry as glucose, adhesives, dextrines and industrial starches.

The United States Testing Company announced that it has officially opened its Chicago Sales Office at 9 West Washington Street, Chicago, Illinois. The main laboratories are located in Hoboken, New Jersey, and the Company has twelve branch laboratories throughout the United States.

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makes seasoning a science

e ability to create and produce a seasoning meet your exact needs, plus the assurance t it is in compliance with Food Additive ws—*IS A SCIENCE AT STANGE.*

The ability and versatility to produce sonings such as Soluble, Liquid, 50-A croground® and Ground for all types of ished products *IS A SCIENCE AT ANGE.*

The technical skill to control the flavor ency of each and every shipment to pre- e specifications—*IS A SCIENCE AT ANGE.*

The talent to create . . . the skill to duce . . . and the flexibility to fit your ds—*IS THE MOTTO AT STANGE.*

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Microground Spice® NDGA Antioxidant® Ground Spice
Cream of Spice Soluble Seasoning®
Peacock Brand Certified Food Color®

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Annual Meeting Announcement

SPECIAL NOTICE

"FOOD SCIENCE AND FEEDING MANKIND",

a panel discussion

will be held at a General Meeting

Tuesday, May 9th

Penn Top Room, Hotel Statler, NYC

8:40 A.M. to 12 Noon

This presentation and discussion will provide a broad, international overview of action that food scientists and scholars are taking to alleviate world food problems. It will encompass major areas of need, and attempt to refute certain misconceptions about the work of food scientists.

The national and international figures making the presentation will report on the amount of work completed, and the even greater area in which work is in progress.

They will show how definitive areas such as protein malnutrition, obesity, and the goal of total nutrition have been studied; where mistakes have been made, and how correct solutions are being put into effect. A report on food science in Russia by an authority who studied the problem firsthand, will reveal many facts which can be compared to our own advanced studies in food technology. What is the best method to apply food technology in underdeveloped countries, and why should certain steps be taken will be outlined by another expert.

Can the human race survive? The technologists will show that, as far as food is concerned, new ideas must be adopted and further reliance on food technologists and scientists will become increasingly important. This is in spite of political repercussions, nuclear weapons and the "population explosion."

In order that all registrants will have the opportunity to attend, THERE WILL BE NO CONCURRENT SESSIONS SCHEDULED.

21st annual meeting • may 7 to 11, 1961 • statler hilton hotel • new york city

PERSONNEL

The Felton Chemical Company, Inc., has appointed **Victor N. Levey** to the post of Research Director. In this position he is responsible for all phases of work concerning the analytical and research laboratories. Mr. Levey received a Bachelor of Science Degree from City College of New York in 1954, and his Master's Degree in Organic Chemistry from Brooklyn College in 1958. Prior to this appointment, Mr. Levey was Technical Director at Tragacanth Importing Corporation. During the past five years he has lectured on Organic Chemistry at the Graduate Division of Brooklyn College.

V. N. Levey

In a recent executive appointment to the national staff of Automatic Canteen Company of America's food service department, **Mr. Alcide E. Bernardin** became manager of food technology. His new responsibilities include: recipe development and control standardization of packaging materials and equipment, standards for preparation, storage and shipping of frozen foods, new products research and coordination among other departments. Formerly production analyst for Wilson & Co., Inc., where he developed a menupack, Mr. Bernardin joined Automatic Canteen last November.

In a recent move designed to concentrate two areas of its research program, Libby, McNeill & Libby created departments of food technology and agricultural research. The change affected eleven IFT Members.

Mr. J. W. Hanley, formerly in new-product planning, has been named Director of Food Technology Research. He will coordinate all research in new-product development, product improvement and manufacturing research, from headquarters at Blue Island, Illinois. **Dr. Gale R. Ammerman** will be associate director of the department, and **C. P. Jingst** will be assistant director. Five managers named are **Dr. R. Franceschini**—new-line products, **T. A. Valvoda**—Manufacturing Research, **T. W. Kendall**—Western Division fruits and vegetables, **D. L. Estes**—citrus, and **M. L. Williams**—Product Research.

Within the new Agricultural Research department, **Dr. O. C. Zoebisch** will assume the position of assistant director, Central, Eastern and Canadian research programs.

Mr. D. McCloskey has been named associate director of research in Hawaii and will be in charge of food technology research as well as agricultural research in the Islands. His assistant director of research will be **W. B. Thomas.**

On March 1st, 1961, **Mr. Joseph H. Hulse** assumed the position of Deputy Director of Research at Maple Leaf Milling Company, Limited. He studied at the University of Manchester and in the Royal College of Science and Technology, Glasgow. Mr. Hulse is past-president of the Canadian Institute of Food Technology, and vice-chairman of the Canadian Committee on Fats and Oils. He served as Associate Editor of FOOD TECHNOLOGY, and is the Canadian representative of the Commonwealth Food Defense Working Group. Prior to his appointment, Mr. Hulse was Head of Food Research in the Defense Research Medical Laboratories, Toronto, Canada.

J. H. Hulse

Appointment of **E. Everett Macdonough, Jr.,** as Manager—Special Accounts of Sterwin Chemicals Inc. has recently been announced. He will be a technical sales-service specialist, working with the research and development departments of major food manufacturers throughout the country. Before joining Sterwin, Mr. Macdonough was associated with Chas. Pfizer & Co., Inc. He received a degree in chemistry from Polytechnic Institute of Brooklyn.

E. E. Macdonough

Other Sterwin changes include appointment of former assistant sales manager **Lowell R. Patton, Jr.,** to the post of manager of the animal health products division and manager of the flour milling and bakery products division, and **F. A. Baldauski** to the post of manager of the food chemicals division.

Ezra Taft Benson, Secretary of Agriculture throughout the Eisenhower administration, has been elected to the Board of Directors of Corn Products Company. He will assume his directorship at the conclusion of his official government duties. In commenting on his appointment, Mr. Benson said, "This new opportunity will enable me to continue my efforts to improve nutritional standards for people everywhere, and to expand markets for farm products throughout the world in association with one of the world's leading food manufacturers." Mr. Benson was born and reared on a farm in Franklin County, Idaho. He is a graduate of Brigham Young University, and did post-graduate work at the University of California.

E. T. Benson

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SITUATIONS WANTED

Unless otherwise specified, particulars on the following individuals may be obtained by writing: Executive Secretary, Institute of Food Technologists, 176 W. Adams St., Chicago 3, Ill. Request by box number.

Recent graduate with Ph.D. degree, major in meats phase of Food Science and minors in Biochemistry and Physiology, desires position in Product Development, Production, Quality Control or Research in connection with meats, meat products or related fields. Also interested in academic position combining teaching and research. Salary $9,000. REPLY BOX 843.

June 1961 Ph.D. graduate in Food Science desires work in teaching or research in Bacteriology and Sanitation of Dairy Products. Experience includes 5 yrs. teaching, research in Detergency and Dairy Bacteriology, Military experience in Veterinary Food Inspection, 1st Lt. QMC. REPLY BOX 844.

Experienced packaging individual desires product development, production, or packaging research position which will allow independent operation. Salary, open. REPLY BOX A-104.

Chemistry graduate desires position in industry research. He has had extensive experience in meats, but would consider a position as chemist working with other products. Salary, $8,000. REPLY BOX A-109.

Position in technical service or as laboratory supervisor desired by 1949 chemistry graduate. Products worked with include sugars, syrups, and confections (250 items). Experience includes quality control, product development, production, packaging, sales and merchandising. Midwest location preferred, but not required. Salary, $14,000. REPLY BOX A-110.

Recent graduate with M.S. in food technology desires position entailing work with meats or poultry, as food technologist or bacteriologist. Desires product development or industry research position. Salary, $6,000. REPLY BOX A-344.

Food technologist or lab technical position desired by individual with eight years quality control experience. She prefers to work with fruits and vegetables, and jams and jellies, but is also interested in cereals and baked goods, sugars and syrups, confections, flavors and colors, and fats and oils. SALARY, $5,700. REPLY BOX A-111.

Recent M.S. in microbiology desires industry research, production, or product development position. Primary area of experience is with algae; however fruits, vegetables, fats, oils, and carbonated, distilled and fermented beverages are also encompassed. Salary, $7,000. REPLY BOX A-346.

Recent M.S. graduate in food technology desires R & D, technical service or production position. Product experience includes meats, dairy, fruits, vegetables, sugars, syrups, poultry and eggs, fats and oils, beverages, and sanitizers. Salary, $8,000. REPLY BOX A-345.

Experienced chemistry graduate desires position working with dairy products or cookies. Her position preference is product development, industry research, and quality control. Salary, $8,000. REPLY BOX A-44.

Industrial research, product development or quality control work with fruits and vegetables interests recent food technology graduate. Salary, $6,000. REPLY BOX A-52.

M.S. with experience in food manufacturing desires position as food technologist, bacteriologist, or in production. Salary, $8,500. REPLY BOX A-318.

Quality control, packaging, and preservation of poultry products interest new Ph.D. in food science. Position preferences also include product development and industry or academic research. Salary, $8,000. REPLY BOX A-639.

1950 graduate with B.S. in food technology desires position in product development, production, technical sales, technical service, packaging, or quality control. Salary, $8,000. REPLY BOX A-2.

Experienced man desires quality control, product development, or production position in the area of vegetables and fruits. Salary, $12,500. REPLY BOX A-8.

1956 graduate with B.Sc. in Home Economics indicates she is interested in a product development or food promotion position. Salary, $6,200. REPLY BOX A-25.

Quality control position with primary emphasis on frozen foods desired by 1955 food technology graduate. Salary, $5,800. REPLY BOX A-30.

Recent chemistry-biology graduate desires laboratory technician or chemist position. Her major experience consists of five years in quality control. Salary, $4,200. REPLY BOX A-62.

1957 chemistry graduate interested in cereals, baked goods, and fats and oils, desires production, technical service or quality control position. Salary, $7,200. REPLY BOX A-74.

Bacteriology graduate desires production or quality control position as food technologist to work with canned foods. Salary, $7,000. REPLY BOX A-77.

Technical service position desired by individual interested in dairy products. Interest range also includes quality control and research. Salary, $8,000. REPLY BOX A-325.

Research position, preferably with industry in the field of citrus products desired. Salary, $8,000. REPLY BOX A-306.

Research food technologist position desired by individual with recent M.S. food technology. Product preference includes meats, vegetables and fruits. Salary, $8,000. REPLY BOX A-311.

Product development, production, or research interest an M.S. food technologist with five years experience in these areas. Salary, $7,500. REPLY BOX A-310.

POSITIONS AVAILABLE

Unless otherwise specified, particulars on the following positions may be obtained by writing, including the company name of your current employer: Executive Secretary, Institute of Food Technologists, 176 West Adams Street, Chicago 3, Illinois. Request by box number.

RESEARCH CHEMIST

To supervise nutritional research activity for major food processor. Ph.D. required with about 5 years post-doctorate experience. Reply giving full particulars including salary requirement to BOX 837.

Position Available—FLAVOR CHEMIST:—Modern growing Flavor Manufacturing concern requires experienced Flavor Chemist to develop new flavor applications and to assist in Quality Control. Ingenuity and initiative needed. Salary commensurate with training and experience. Opportunities for growth unlimited. Please submit résumé with salary requirements. All replies held confidential and all will be answered. REPLY BOX 838.

WANTED: Production man for food plant. Knowledge of pickles and sauerkraut desirable but not essential. Mid-West location. Salary open. Reply in detail to BOX 839.

SALES OPPORTUNITY

This is a small ad offering a big opportunity for outstanding salesman with experience contacting food and beverage manufacturers with raw materials. Salary open; full company paid benefits; profit sharing plan. Reply with salary requirements to BOX 840.

WANTED: College graduate, major in Chemistry or Food Technology for work in southeastern pickle plant as head of Quality Control and Assistant Plant Manager. Production background preferred, but pickle experience not required. Established company. Reply should include education, age, experience and present salary. BOX 841.

TECHNICAL SALESMAN

Opportunity with expanding, financially sound food emulsifier manufacturer. Old established company moving into modern new plant spring 1961. Position requires experience in technical sales support or sales. Assignments demand negotiation and selling at highest policy-making level. Location will be New York City. Salary open. Address replies to BOX 842.

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Senior flavor chemist. East coast location. Ten years experience and B.S. in chemistry or equivalent, to create and improve flavor compounds in the essential oil field. REPLY BOX P-604.

Food technologist. Eastern location. B.S. degree and no experience required, to conduct research investigations for product development. REPLY BOX P-606.

Food technologist. Midwest location. B.S. in food technology or chemistry with 5–8 years experience. Considerable travel to assist customers with product applications. REPLY BOX P-633.

Assistant Director of Research. Midwest location. Degree in chemistry or food technology with ten years experience in developing food products, and a history of independent operation desired. Position includes new product development in dairy, ice cream, and food vending fields. REPLY BOX P-733.

Microbiologist. Midwest location. Two years experience in field related to bacteriology, which should have been college major. Applicants must be capable of assuming responsibility in carrying out practical and fundamental investigations on vinegar production and physiology of Acetobacter. REPLY BOX P-723.

Quality Control Manager Trainee. Eastern location. Minimum educational requirement is B.A. or B.S. in food technology, with experience in bacteriology, dairy, egg and bakery products desirable. Position will lead to supervisory capacity. REPLY BOX P-724.

Product Development Supervisor. Midwest location. Two to four years experience in product development, food preservation and packaging desired. Scientific degree required for work entailing investigative work on formulations and preservation of foods. REPLY BOX P-731.

Manager—Statistical Quality Control. A scientific degree and two to four years of statistical quality control work required. Individual selected will develop and maintain a statistical quality control program. REPLY BOX P-732.

Chemist. Midwest location. Degree in food technology, biochemistry or related field. No experience required. Duties entail product development and applied research on nutritional specialty products. REPLY BOX P-734.

Food technologist. Midwest location. Technical degree required plus 0-2 years experience. Maintenance of quality and development of new products including coffee, vegetable oil products, glass packs, etc. A non-routine position necessitating willingness to travel. REPLY BOX P-725.

Dairy technologist. Midwest location. Technical degree required plus at least two years experience in grading. Position entails cheese grading and approving purchases of cheese and other dairy products. Individual selected must be willing to travel. REPLY BOX P-726.

Development Bacteriologist. Midwest location. B.S. or M.S. in bacteriology plus one year of industrial bacteriology work on meats. Position entails bacteriological analyses, specifications, and development research. REPLY BOX P-729.

Technologist. Midwest location. B.S./M.S. in chemistry and/or bacteriology for quality control position involving chemical and bacteriological control of raw ingredients, processing parameters and finished product. REPLY BOX P-730.

Instructor in research. Midwest location. Background in chemistry, bacteriology, or statistics necessary to work in quantity food research program in a College of Home Economics. REPLY BOX P-701.

Food Technologist. Midwest location. B.S. in food technology or related areas required. Position entails food product development with all commodities. REPLY BOX P-712.

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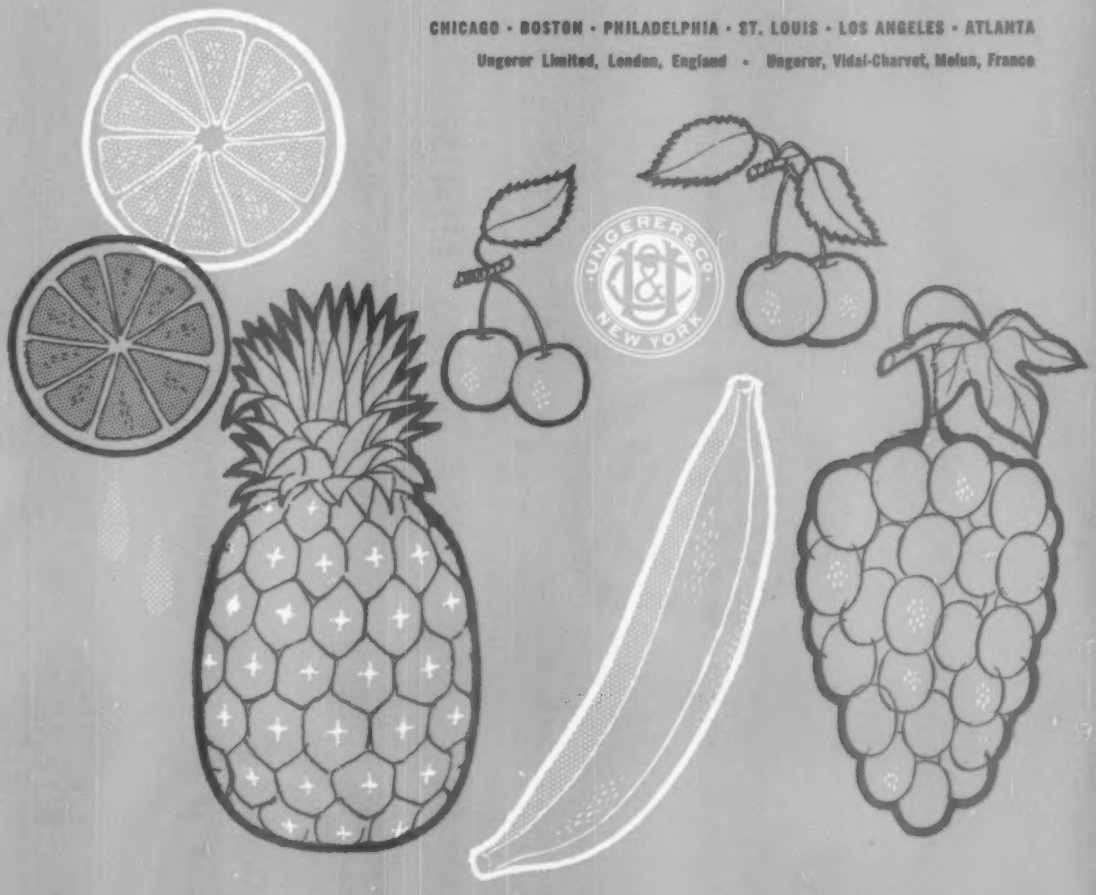

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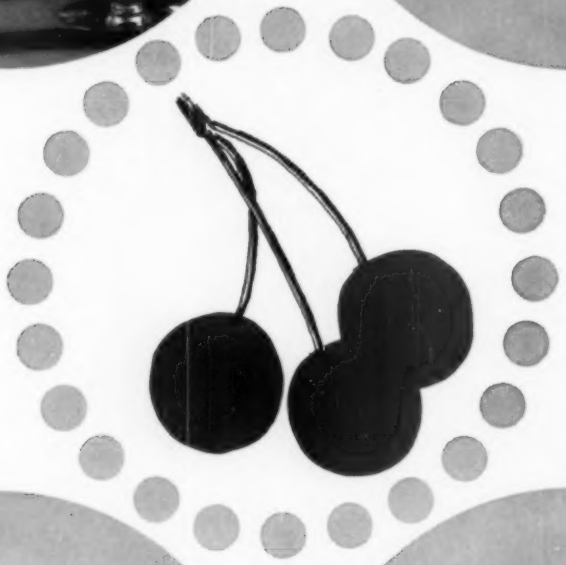

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